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New Era in the Drug Delivery System: Formulation and Evaluation of Microsponge of Itraconazole

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Abstract: ***Microsponges are polymeric delivery systems composed of porous microspheres. They are tiny sponge-like spherical particles with a large porous surface. Microsponge is recent novel technique for control release and target specific drug delivery system. They are desire to deliver API efficiently at the minimum dose and also to enhance stability, reduce side effect and modify drug release. Itraconazole Microsponges weresuccessfully prepared by a quasi-emulsion solvent diffusion method by using polymers such asethyl cellulose,polyvinyl alcohol,triethyl citrate (TEC20-40%) as plasticizer.Itraconazole loaded microsponge was prepared in a ratio of 1:1,1:2,1;3,1;4,1:5,1:6,1:7,1:8,1:9 and porous spherical & stable, microspongeare obtained in the range of 40% to 45% and above.This microsponge evaluated by SEM, Particle size analysis, Entrapment efficiency, DSC,FTIR,IR spectroscopy.This studyshowed that microsponge werefound sphericaland porous surface structure, more stable, inertand drug is molecularly dispersed in the formulation.***

Keywords: API: Active pharmaceutical ingredient, DSC: Differential scanning calorimetry, IR: Infrared,FTIR: Fourier transforms infrared, SEM: Scanning electron microscopy

1. Introduction

Microsponge technology was a firstly developed by Won in 1987 and original patents were assigned to advanced polymer system. They are tiny sponge-like spherical particles with a large porous surface. Microsponge is recent novel technique for control release and target specific drug delivery system. They are desire to deliver active pharmaceutical ingredientefficiently at the minimum dose and also to enhance stability, reduce side effect and modify drug release. Polymericbased microsponge uniquely overcome problems associate with above technologies. Microspongesare extremely small, inert, imperishable spheres that do not pass through the skin. Spherical particles composed of clusters of even tinier Microsponges are capable of holding four times their weight in skin secretions. These products are typically presented to the consumer in conventional forms like creams, gels or lotions and they contain a relatively high concentration of active ingredients. The present study is an attempt to formulate microsponge novel drug delivery system of Itraconazole, an orally administered anti-fungal drug with a view of improving its oral bioavailability and giving a prolonged release of drug. Itraconazole is a drug which is absorbed from the gastrointestinal tract (GIT) and has a longer half-life but eliminated quickly from the blood circulation, so it required frequent dosing. To avoid this drawback, the oral controlled release formulations were developed in an attempt to release the drug slowly into the GIT and to maintain an effective drug concentration in the serum for longer period of time. The word new or novel in the relation to drug delivery system is a search for something out of necessity. An appropriately designed sustained or controlled release drug delivery system can be major advance to words solving the problem associated with the existing drug delivery system. Oral controlled release multiple unit dosage forms such as beads, pellets and microspheres and microsponges are becoming more popular than single unit dosage forms due to several inherent advantages. Microsponges are play role to novel drug delivery system

Recently their use is also being investigated for oral drug delivery.Microsponge consists of non-collapsible structures with porous surface through which active ingredients are released in a controlled manner. Their characteristic feature is the capacity to adsorb or “load” a high degree of active materials into the particle and on to its surface.To prevents excessive accumulation of ingredients within the epidermis and the dermis. Controlled release of drug on to epidermis does not enter the systemic circulation in significant amounts.Active ingredients that are entrapped in microsponge can then be incorporated into many products such as creams, gels, powders, lotions and soaps.

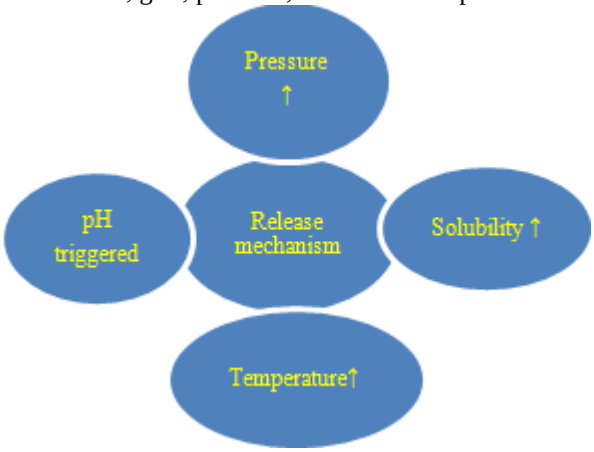


Figure 1: Drug release mechanism of microsponges

Characteristics of microsponges: [4, 5, 7]
They are compatible with most vehicles and ingredients.

Microsponge formulations are stable over range of pH 1 to 11.
It is stable at the temperature up to 130º c.
They are self-sterilizing as their average pore size is 0.25µm where bacteria cannot penetrate.
Microsponge formulations have high entrapment efficiency up to 50 to 60%.

Characteristics of materials entrapped in microsponges: [10,23,15,17]

Most of liquid or soluble ingredients can be entrapped in the microsponge.
They should be completely miscible in monomer& should be inert to monomers.
They should be stable in contact with polymerization catalyst.
They should be water immiscible or at most only slightly soluble.

A number of different substances both biodegradable as well as non-biodegradable have been investigated for the preparation of microsponges. These materials include the polymers of natural and synthetic origin and also modified natural substances. Synthetic polymers employed as carrier materials are methyl methacrylate, acrolein, lactide, glycolide and their copolymers, ethylene vinyl acetate copolymer, polyanhydrides, etc. Following are the methods of preparation of microsponge.

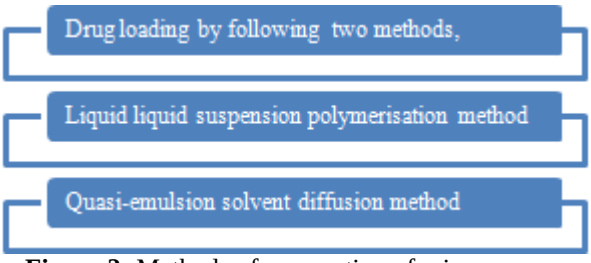


Figure 2: Methods of preparation of microsponges

Mechanism of action: These are the chemical agents who are used in the treatment of infections caused by fungi are called as antifungal agent. Azole derivative act by damaging the fungus cell membrane. All azole act by inhibiting of 14 the C-P 450 sterol α- demethylase inhibition of the 14 α-demethylase results in accumulation in the fungal cell membrane of sterols still bearing a 14 α- methyl group. This results in permeability changes, leaky membrane & malfunctions of membrane imbedded proteins. These effects taken together lead to inhibition of fungal cell growth not the fungal growth so azole is the fungiststic. Itraconazole is an imidazole / triazole type antifungal agent. It is a highly selective inhibitor of fungal cytochrome p-450sterol C-14 α-demethylation via the inhibition of enzyme cytochrome P-450 14 -a demethylase. This enzyme converts lansterol to ergo sterol& required in fungal cell wall synthesis.

2. Material and Method

The drug Itraconazole & Polymers was procured/gifted Emcure by pharmaceutical companies and are listed in Table 1

Table 1: List of active ingredient and polymers used in the work

Materials	Grade / Specification	Manufacturer / Supplier
Itraconazole	Research Grade	Nifty Lab Pvt Ltd, Hyderabad
Eudrajit RS100	Research Grade	LobaChemiePvt.Ltd.Mumbai
Triethyl Citrate	Analytical Reagent	LobaChemiePvt.Ltd.Mumbai
Polyvinyl Alcohol	Analytical Reagent	LobaChemiePvt.Ltd.Mumbai
Ethanol	Analytical Reagent	LobaChemiePvt.Ltd.Mumbai
Methanol	Analytical Reagent	LobaChemiePvt.Ltd.Mumbai
Ethyl cellulose	Research Grade	LobaChemiePvt.Ltd.Mumbai
Dichloromethane	Analytical Reagent	LobaChemiePvt.Ltd.Mumbai
Eudrajit RSPO	Research Grade	Emcurepharma, Pune
Eudrajit RLPO	Research Grade	Emcurepharma, Pune

Table 2: List of equipment and manufacturers

Name of Equipments	Manufacturer	Model No.
Magnetic stirrer	REMI	1MLH
Analytical balance	AFcoset	FX - 300
Ultrasonicator	LABLINE	1-5L-50
UV- visible spectroscopy	SHIMATDZU	UV-100
Stability testing chamber	THERMOLAB	
Dissolution apparatus	ELECTROLAB	TDT 08L
Digital pH meter	-	-
Fourier transform Infrared spectrophotometer	JASCO	4600
Scanning electron microscope	JEOL	SM-6360
Differential scanning calorimeter	METTLER	
Hot Air Oven	YORCO Scientific Industry	YSI-431

2.1 Preformulation study of drug

2.1.1 Analytical Parameters

a) Determination of λ max: The λmax was obtained at 263 nm in methanol and 257nm in 0.1N HCl shown in Figure no.3

Figure 3: λ max ofItraconazole

b) Standard curves ofItraconazole:

The calibration curve of Itraconazole was obtained by dissolving the drug in mixture of methanol and dichloromethane, mixture of methanol and dichloromethane was used as co-solvent. The graph of absorbance vs. concentration for Itraconazole was found to be linear in the concentration range 1-10 µg/ml in methanol and 1-10 µg/ml, in 0.1N HCl. The slope and correlation coefficient of the calibration curve were found to be 0.992 in methanol and 0.999 respectively as shown in Table No.6. The drug obeys Beer-Lambert's law in this concentration range.

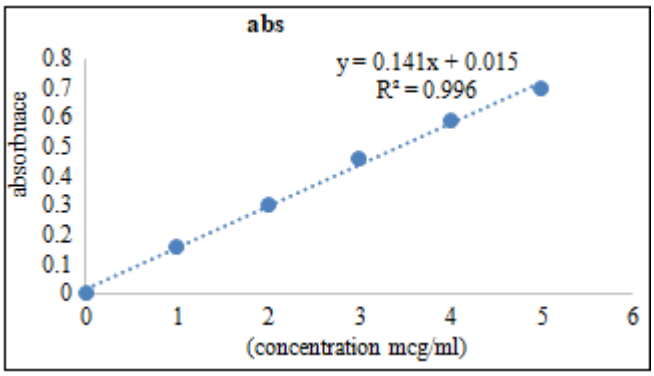


Figure 4: Calibration curve of Itraconazole

2.2 Preliminary study

a) Appearance and color

The drug Itraconazole was examined for its organoleptic properties like color and appearance. The sample was observed white crystalline powder.

b) Melting point

The melting point of Itraconazole was determined by capillary method using Thiel's tube. Melting point is a first indication of purity of sample. The melting point was recorded in range of 166-170^{o}C, which was in the range of reported melting point and shown in Table no.3.

Table 3: Melting point of Itraconazole

Name	Melting pointobserved	Melting pointreported
Itraconazole	166^{0} C.	166-170^{0} C.

2.3 Identification of drug

a) Infrared spectroscopy

IR absorption spectrum of Itraconazole was recorded by potassium bromide dispersion technique using JASCO(4600) FT-IR spectrophotometer as shown in Figure 5. Characteristic peak at 1697.12cm^{-1}corresponds to C=N stretching,2820.44cm^{-1}due to O-H Stretching,669.94cm^{-1} due to C-X Stretching, 1550.63cm^{-1}due toN-H stretching,1270.84cm^{-1} due to C-O stretching.

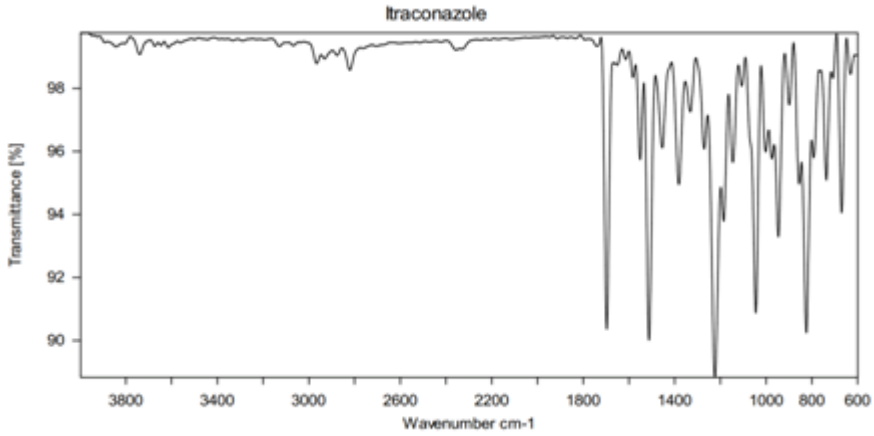


Figure 5: IR absorption spectrum of Itraconazole

b) Differential scanning calorimetry

DSC study was performed for drug. DSC measurements were performed on a METTLER STAR 9.01 instrument at 40^{o}C to 300^{o}C using a sealed aluminum pan. The DSC Thermogram of Itraconazole showed characteristic sharp endothermic peaks at 169^{o}C as shown in Figure 6. This indicates melting point of the drug.

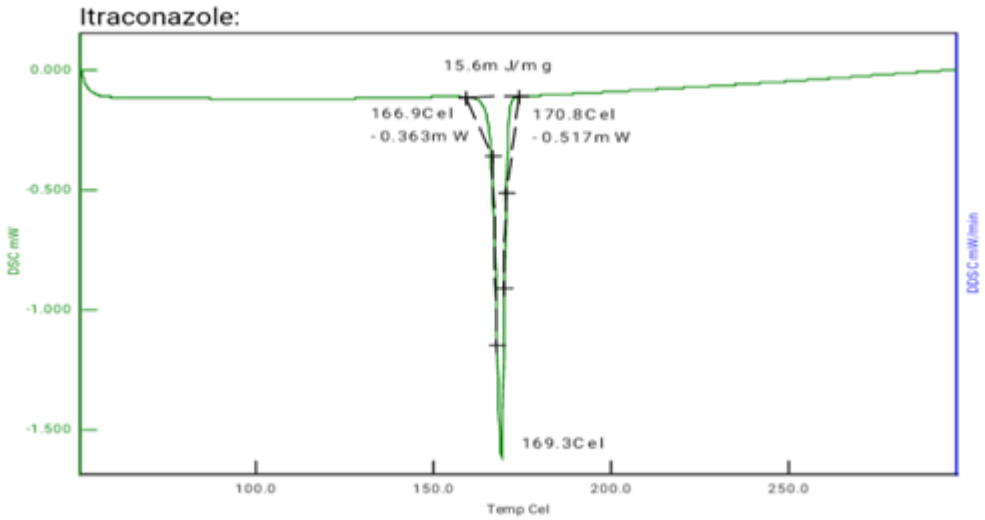


Figure 6: DSC of Itraconazole

2.4 Identification of polymer

a) Melting point

In Preformulation study of polymers, the melting point for ethyl cellulose was 161- 166˚C and PVAwas 204-210^{o}C recorded shown in Table No.11.

b) FTIR Study of polymers

FTIR spectrum of ethyl cellulose were recorded on BRUKER FT-IR spectrophotometer over the region spectra's were shown in Figure 7 respectively. In FT-IR spectra for ethyl cellulose,the characteristics peak at 1048.53cm^{-1}due to C-O,1444.85cm^{-1}due to C-O-H Bending,2970.68cm^{1} due to C-H Stretching,915.45cm^{-1}due to =CH Bending , 1371.93cm^{-1}due to ALKENE C=CH. In Figure.7 and table no.4.respectively

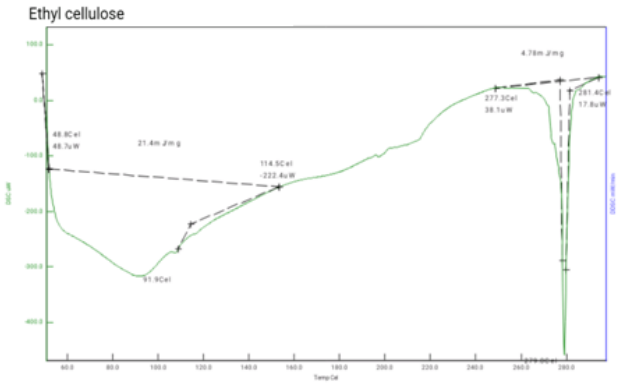


Figure 7: FTIRethyl cellulose

Table 4: FTIR spectrum of ethyl cellulose

Functional group	Vibrational frequencies (cm^{-1}) Observed	Vibrational frequencies (cm^{-1}) reported
C-H Stretching	2970.68	3000-2850
C-O-H Bending	1444.85	1440-1220
C-O Stretching	1048.53	1260-1000
=CH Bending	915.45	1000- 650
Alkene C=CH	1371.93	1380-1370

c) Differential Scanning Calorimetry:

DSC studies for ethyl cellulose and PVA were performed. The DSC Thermogram showed two characteristic peaks at 289^{o}C and 230^{o}C respectively as shown in Figure 8, and Figure 9. Respectively.

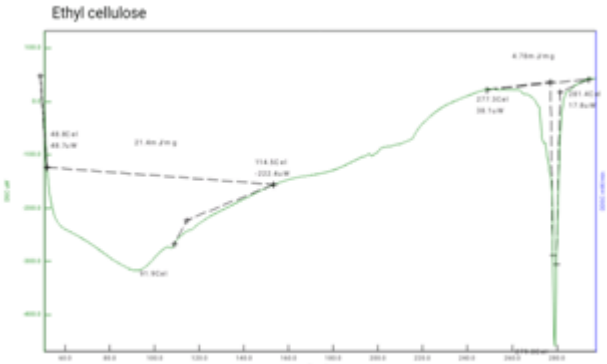


Figure 8: DSC of ethyl cellulose

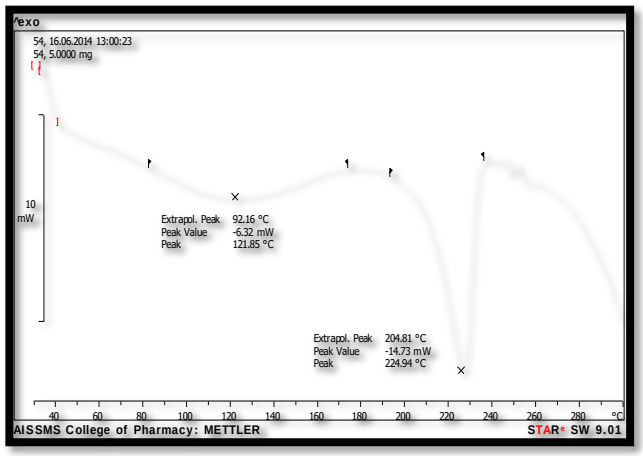


Figure 9: DSC of PVA

2.5 Formulation and Evaluation

a) Formulation of Itraconazole microsponge by quasi emulsion method: (2 step process)

Itraconazole microsponges were prepared by a quasi-emulsion solvent diffusion method, which was carried out in two step using, an internal phase containing required amount of polymer such as ethyl cellulose weighted accurately and dissolved in 20ml methanol and dichloromethane (1:1).The drug Itraconazole was slowly added to the polymer solution

and dissolved under ultra-sonication at 35ºC. The internal phase made up of different drug –polymer ratios and plasticizer such as tri ethyl citrate (TEC20-40%) was added in order to aid the plasticity. External phase containing polyvinyl alcohol dissolved in distilled water at temperature 60ºC was prepared and allowed to cool room temperature. The internal phase at a room was added drop wise with the help of syringe (1ml) and needle into external phase withcontinuous stirring at 1000 rpm for 3 hrs. Until complete diffusion of DCM and Methanol in internal phase takes place, after complete diffusion of DCM and Methanol the mixture was filtered to separate the microsponges. The prepared microsponges were washed and dried temperature at35°C for 24 hrs

Figure 10: Prepared microsponges

Formulation batches

Table 5: Formulation batches of Itraconazole microsponges

Formulation Code	Drug: polymer (mg)	Ethyl cellulose (mg)	Triethyl citrate (ml)	Polyvinyl Alcohol (mg)	External phase volume (ml)	Internal Phase volume (ml)
F1	(1:1)	100	4	500	100	20
F2	(1:2)	200	4	500	100	20
F3	(1:3)	300	4	500	100	20
F4	(1:4)	400	4	500	100	20
F5	(1:5)	500	4	500	100	20
F6	(1:6)	600	4	500	100	20
F7	(1:7)	700	4	500	100	20
F8	(1:8)	800	4	500	100	20
F9	(1:9)	900	4	500	100	20

b) Evaluation of microsponges

Micromeritics Properties of Prepared Microsponges:[28,29]

Table 6: Observation table of Micromeritics properties of microsponges

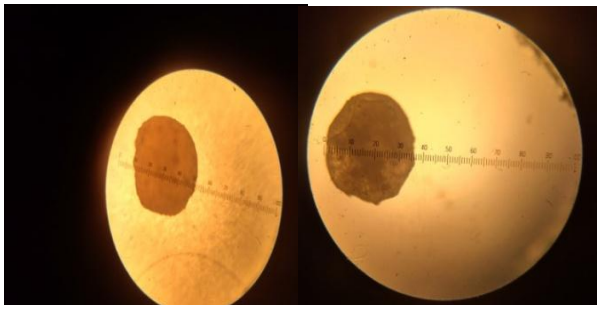
Batch	Bulk density gm/cm^3	Tapped density gm/cm^3	Hausner's ratio	Carr's/ Compressibility index %	Angle of repose (degree)
F4	0.453±0.001	0.474±0.003	1.053±0.025	9.36±0.150	26.8±0.089
F5	0.424±0.001	0.464±0.001	1.076±0.015	6.45±0.064	23.96±0.597
F6	0.440±0.001	0.454±0.002	1.046±0.015	9.45±0.064	26.36±0.12
F7	0.285±0.038	0.315±0.003	1.096±0.025	7.63±0.563	22.93±0.19
F8	0.489±0.007	0.575±0.002	1.12±0.025	8.73±0.387	26.81±0.032
F9	0.237±0.015	0.445±0.001	1.13±0.026	12.40±0.455	29.11±0.025

c) Particle size determination:[25]

D (mean) =∑n d / ∑n where, n= no. of microsponges observed, d= mean size range.

Table 7: Observation table of particle size of Itraconazole Microsponges

Batch.	Particle size(µm)
F4	172
F5	175
F6	236
F7	283
F8	290
F9	293



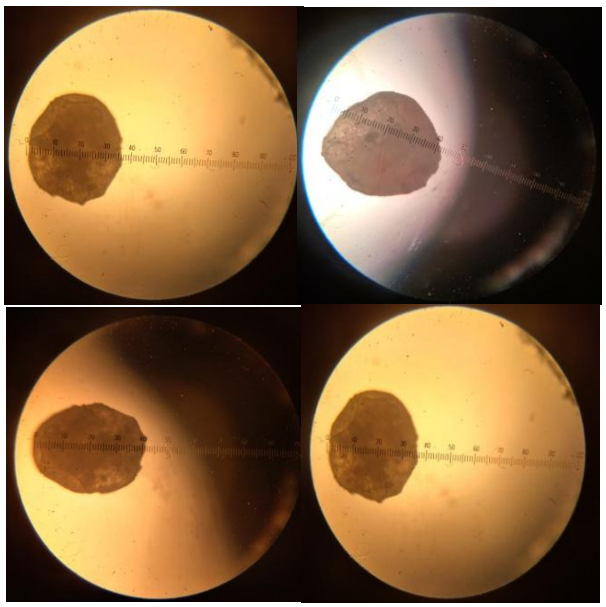


Figure 11: Photographs of determination of practical sizeofItraconazole microspongeformulation F4–F9 by using optical microscope

d) Percentage yield:

$$\text{Percent yield} = \frac{\text{Amount of microsponges obtained} \quad X100}{\text{Theoretical amount (drug + polymer amount)}}$$

Table 8: Percentage yield for microsponges

Batch	% Percentage yield
F1	-
F2	-
F3	-
F4	40%
F5	43%
F6	42%
F7	43%
F8	44%
F9	45%

The prepared microsponges were evaluated for the following parameters:

e) Drug content:[11,12]

Microsponges equivalent to 100 mg of Itraconazole were dispersed and made up to the mark in the 100 ml volumetric flask with 30 ml 0.1 N HCl buffer, sonication in ultrasonicator to extract the drug. After filtration through Whatman filter paper, from this filtrate 1ml of aliquot was diluted suitably get 10 µg/ml concentrations and drug content was analyzed by UV spectrophotometer at 257nm against blank. . Percentage drug content

$$\text{Drug content} = \frac{\text{Actual drug content}}{\text{Theoretical drug content}} X\,100$$

Table 9: Observation table of Drug content

Batch no.	Microsponge(mg)	Percentage of drug content
F4	100	95%
F5	100	51%
F6	100	96%
F7	100	48%
F8	100	60%
F9	100	70%

f) Determination of drug entrapment efficiency:[30, 31]

The entrapment of drug loaded microsponges was determined by dispersing 100 mg microsponges in 30 ml 0.1 N HCl allow to keep overnight. After filtration through Whatman filter paper, from this filtrate 1ml of aliquot was diluted suitably get 10 µg/ml concentration and drug content was analyzed by UV spectrophotometer at 257nm against blank. Percentage drug content and drug entrapment efficacy was calculated by following formula:

$$\textbf{g)Entrapment efficiency of drug} = \frac{\text{Actual drug content} \quad X\,100}{\text{Theoretical drug content}}$$

Table 10: %Entrapment efficiency of Itraconazole microsponges

Batch	Entrapment efficiency (%)
F1	-
F2	-
F3	-
F4	95.5%
F5	51.2%
F6	96.3%
F7	48.6%
F8	60.1%
F9	70.4%

h) Scanning electron microscopy (SEM):[30,32]

Morphology and surface characteristics were studied by Scanning Electron Microscopy. The samples for the SEM analysis were prepared by sprinkling the microsponges on one side of the double adhesive stub.

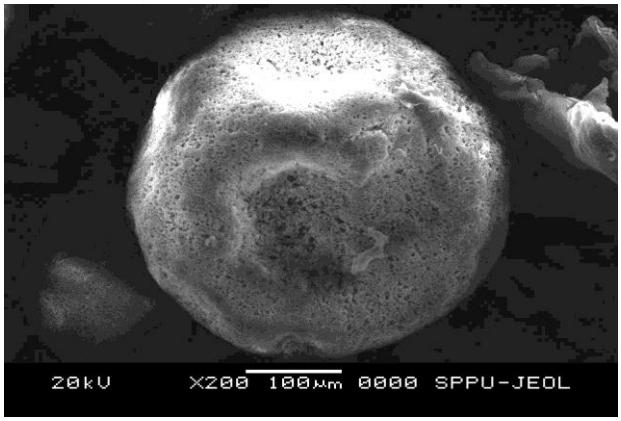


Figure 12: SEM Photographs of Itraconazole microsponge formulation

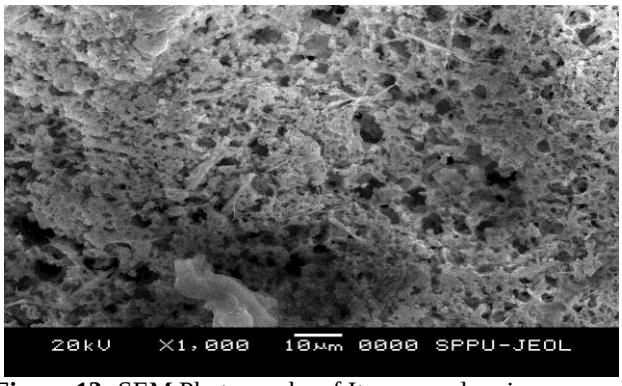


Figure 13: SEM Photographs of Itraconazole microsponge formulation

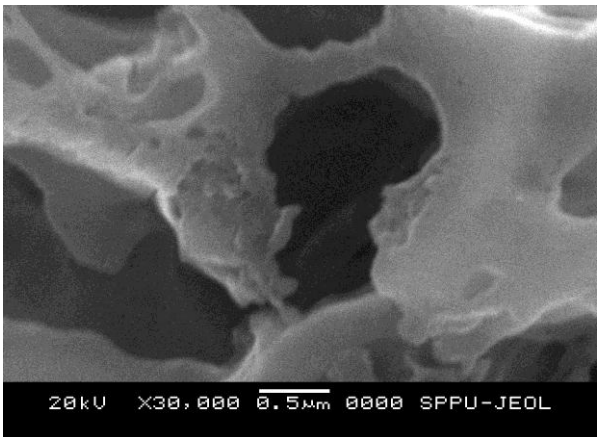


Figure 14: SEM Photographs of Itraconazole microsponge formulation

i) Differential scanning calorimetry (DSC):[30, 31, 12]DSC study was carried out for formulation on a METTLER Toledo (823E). Indium/zinc standards were used to calibrate the DSC temperature and enthalpy scale. The sample were sealed in aluminum pans and heated at a constant rate 20 ^{o}C /min over a temperature range of 40-250^{o}C. DSC thermo gram for formulation showed a shallow and broad endothermic peak at 157.7^{0}C, while DSC thermogram of

Itraconazole showed a sharp endothermic peak at 169^{o}Cit is indicates drug is molecularly dispersed in the polymers.

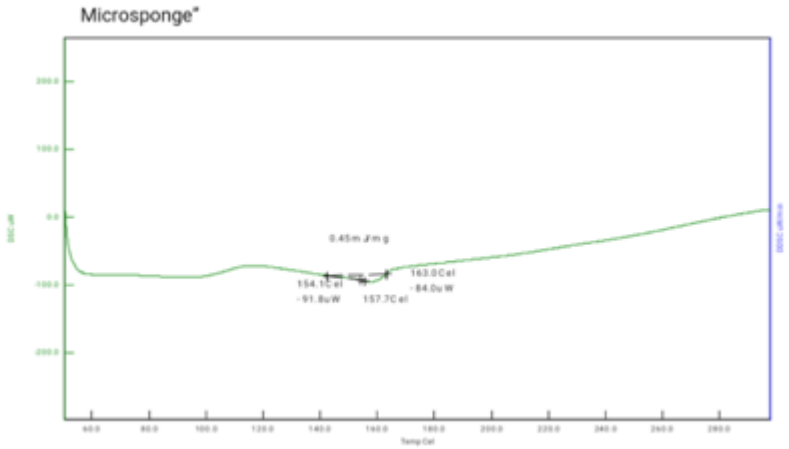


Figure 15: DSC Thermogram for microsponge formulation

j) FTIR of formulation batch:[19,15-20]FTIR study of formulation was carried out on FTIR apparatus JASCO at Pune University, Physics Department. The optimized formulation was considered for FTIR analysis. The sample was scanned over a range of 4000-400 cm^{-1} using Fourier transformer infrared spectrophotometer. Spectra's were analyzed for drug polymer interactions.

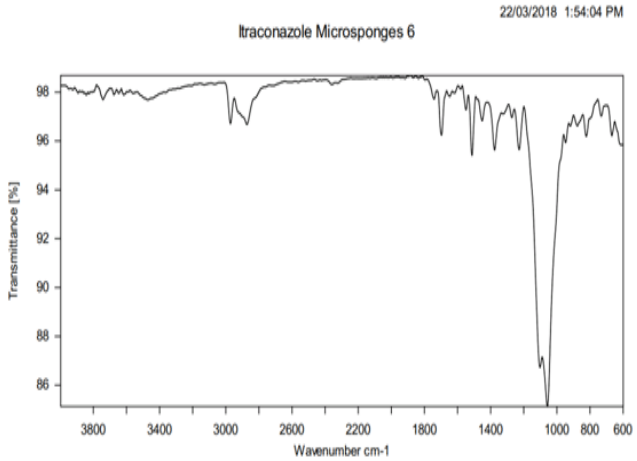


Figure 16: FTIR of microsponges

Table 11: Interpretation of infrared spectrum values

Functional group	Vibrational frequencies (cm^{-1})	Reported
O-H stretching	2971.80	3000-2800
C-Cl Stretching	822	500-100
C-O stretching	1227.59	1260-1000

k) *In- Vitro* drug release studies:[30-32]*In-vitro* dissolution studies carried out using USP XXIVdissolution assembly (basket type, Electro lab TDT-08L) in 900 ml of 0.1N HCl, accurately weighted samples of the microsponges were packed in muslin cloth and placed into basket and immersed into dissolution medium and a stirring rate of 50 rpm and temperature of 37±0.50C. Drug release monitored for 6h. A sample (5 ml) withdrawn at regular time intervals and sink conditions was maintained by replacing an equal amount of fresh dissolution medium. The samples wereanalyzed spectrophotometric ally (ELICO) at a wavelength of 257 nm.1 ml aliquots were withdrawn diluted up to 10 ml with 0.1 N HCl and replaced with an equal volume of fresh dissolution medium. After suitable dissolution, the amount of drug released was calculated using standard calibration.Percentage cumulative release versus time.The dissolution data subjected to various release models, namely, Zero order, first order, Higuchi and Korsemeyer-Peppas.

Table 12: Model fitting data for F4 to F9 formulation

Microsponge batches	N	k	Best fit model	R value
F4	1.1310	0.1069	Hixson – Crowell	0.9923
F5	0.9391	0.4533	Korsemeyer-Peppas	0.9938
F6	0.8089	1.0699	Korsemeyer-Peppas	0.9820
F7	0.7212	1.7711	Korsemeyer-Peppas	0.9856
F8	0.6561	2.5046	Korsemeyer-Peppas	0.9877
F9	0.5969	3.4812	Korsemeyer-Peppas	0.9890

Table 13: Model fitting data for F4 to F9 formulation

Release exponent (n)	Drug transport mechanism
0.5	Fickian diffusion
0.5<n<1.0	Anomalous transport
1.0	Case II transport
Higher than 1	Super case II transport

Table 14: *In-vitro* cumulative % drug release form F6 formulation

Time (min)	*Absorbance*	*% Cumulative Drug release*
0	0.0	0.0
30	0.03	13.52
60	0.045	22.49
90	0.056	28.48
120	0.065	34.48
180	0.076	40.49
240	0.085	46.51
300	0.096	52.53
360	0.105	53.78
420	0.110	54.43
480	0.115	55.09

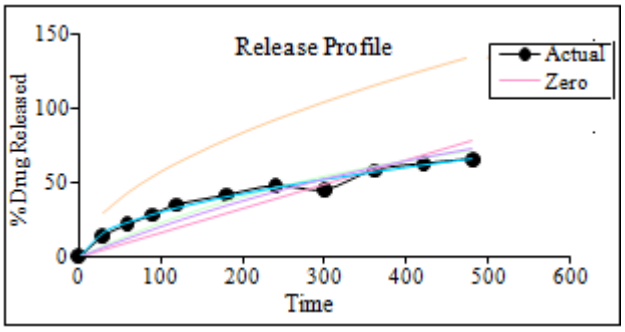


Figure 17: Model fitting curves for formulation

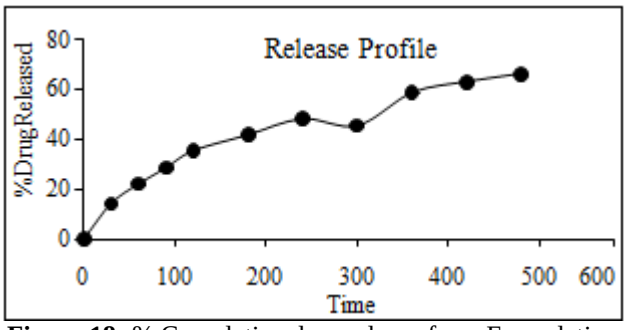


Figure 18: % Cumulative drug release from Formulation

2.6 Analysis of release kinetics

To analyze the mechanism for the release and release rate kinetic of the dosage form,the data obtained was fitted in to Zero order, First order, Higuchi matrix and Korsemeyer-Peppas model. By comparing the R2 values obtained, the best-fit model was selected.

- For Batch F4, the diffusion exponent (n) value was found to be 1.1310 which suggest that the drug release is Super case II transport and best fit model is Hixson – Crowell model which indicatessustained release of the drug
- For Batch F5, the diffusion exponent (n) value was found to be 0.9391 which suggest that the Fick's law of diffusion was not followed that is anomalous transport. From the result, the best fit model for formulation F5 is Korsemeyer-Peppas model.
- In case of Batch F6, the diffusion exponent (n) value was 0.8089 indicates that the anomalous transport or non-Fickian type drug diffusion takes place. From the result, the best fit model for formulation F6 is Korsemeyer-Peppas.
- For Batch F7, the diffusion exponent (n) value was found to be 0.7212 indicates that the anomalous transport or non-Fickian type drug diffusion takes place. From the result, the best fit model for formulation F6 is Korsemeyer-Peppas model.
- For Batch F8, the diffusion exponent (n) value was found to be 0.6561 indicates that the anomalous transport or non-Fickian type drug diffusion takes place. From the result, the best fit model for formulation F6 is Korsemeyer-Peppas model.
- For Batch F9, the diffusion exponent (n) value was found to be 0.5969 indicates that the Fickian diffusion. From the result, the best fit model for formulation F6 is Korsemeyer-Peppas model.

In the area of pharmaceutics, the usual practice is to understand the release kinetics of a drug through a polymer matrix using an empirical relationship proposed by Ritger and Peppas if n= 0.5, the drug diffuses andreleases through the polymer matrix following Fickian diffusion. Ifn> 0.5, anomalous or non-Fickian type drug diffusion takes place and if,n= 1, it indicates a non-Fickian or Case II release kinetics. The values ranging between 0.5 and 1.0 are attributed to anomalous type diffusive transport

2.7 X-Ray diffraction (X-RD) studies:[23]

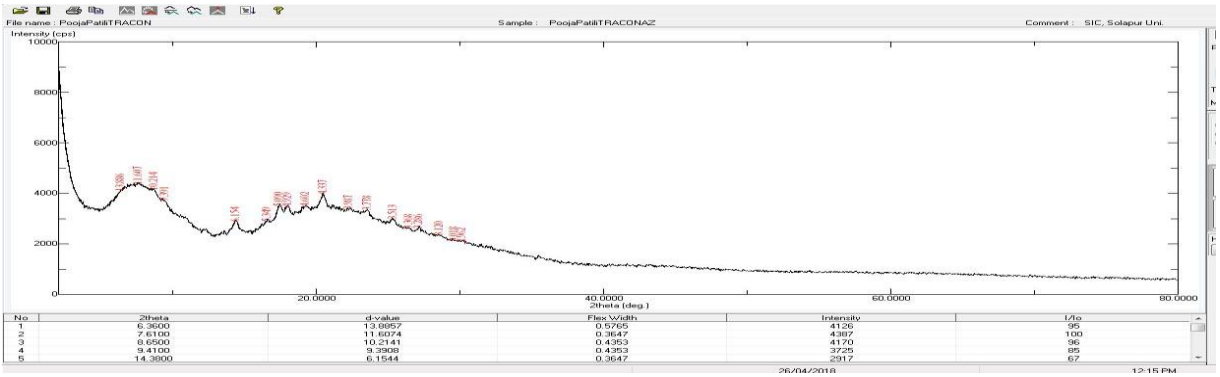


Figure 19: X-Ray diffraction for formulation

The X-Ray powder Diffraction patterns was obtained at room temperature using a X-ray diffractometer with a Cu as anode material and graphite as monochromatic, operated voltage of 35 kV, current 20 mA. The sample were analyzed in 2 θ angle range of 10^{o}-$20^{o}$the process parameters were set as scan step size of 0.02^{o} (2 θ), scan time is 0 to 60 second.In above XRD pattern, intense peaks between 2θ of 10 to 20^{o} were observed shows crystalline nature of microsponges.

3. Discussion and Summary

3.1 Evaluation of Microsponges

a) Micromeritics properties: All the micromeritics properties of all formulations were evaluated such as bulk density, tapped density, Hausner's ratio, compressibility index and angle of repose.Bulk density for all formulations were found in the range of 0.4536 to 0.237 gm/cm^{3}.Where as tapped density values for all formulation were found to be 0.474 to 0.445 gm/cm^{3}. Hausner's ratio, Carr's index and angle of repose were found in the range 1.053 to 1.13, 9.36

to 12.40 % and 26.8 to 29.11°respectively as shown in table No.6. The values for Angle of repose, Hausner's ratio and compressibility index were found to be in good correlation indicating that all formulation possess excellent flow property which confirmed free flowing nature of the microsponges.

b) Percentage yield: Percent yield of the microsponges was calculated, the yield of Itraconazole loaded microsponges was obtained in the range of 40% to 45%. Observation table showing percentage yield of F4-F9 batch was given in table no.08

c) Formulation and development: Itraconazole microsponges were successfully prepared by quasi emulsion solvent diffusion method.

d) Entrapment efficiency: The actual amount of Itraconazole present in the different formulations of microsponges was determined by measuring entrapment efficiency.Drug entrapment efficiency (% DEE) was found to be in the range between 48 to 95% as shown in Table No.10.

e) Particle size analysis: During particle size analysis, it was observed that the size of microsponges depends upon the ratio of polymer and extent ofemulsifying agent. Results of particle size of Itraconazolemicrosponges were presented in Figure 11and Table No.7. .It was observed that the average size the microsponges range from 172 µm to 293 µm. Thus the data showed a systematic dependence the ratio of ethyl cellulose and PVA used while formulating the microsponges.

f) *In- Vitro* drug release study: The *in-vitro* release data obtained for formulation F4 to F9 was shown in Table 12 and Figure No.31 to 43 respectively. The cumulative percent drug release after 8 hourswas found to be 91.62, 76.69, 64.66, 62.84, 60.18, and 52.68for formulation F4 to F9 respectively. The results obtained shows that the drug release was lowest 52.68% for F9 formulation while high drug release 91.62% for F4 formulation and Model fitting and Kinetic assessment of drug release from microsponges shown in Table No.12. F4 batch showed n value 1.1301, if n=1 indicates non Fickian or case II release kinetics. Korsemeyer Peppas model best described the sustained release for F5 formulation and the diffusion exponent (n) value was found to be 0.9391 suggesting that the Fick's law of diffusion was not followed. From the results, the best fit model for formulation, F4 is Hixson Crowell and F5 to F9 is Peppas model. The values of n varied within 0.680 to 0.888, indicated anomalous type diffusive transport F6, F7, F8 shows n value0.8089, 0.7212, 0.6561, this indicates that the values of n varied within 0.680 to 0.888, indicated anomalous type diffusive transport (Table No.14).F9 batch shows n value 0.5969, the values ranging between 0.5-1.0 are attribute to anomalous type diffusive transport.

g) Scanning electron microscopy: SEM micrographs showed that microsponges were spherical, porous surface structure of microsponge, shown in Figure 12, 13 and 14.

h) FTIR: FTIR spectrum for microsponge formulation (fig.16) showed shift and disappearance of some characteristic peaks of the drug. It suggests that drug is molecularly dispersed in the formulation.

i) Differential scanning calorimetry (DSC): The DSC curve of Itraconazole showed sharp melting endothermic peaks at 169°C correspondingto melting point ofItraconazole as shown in Figure No. 15. The disappearance of melting point endothermic peak of drug in microsponges indicates that the drug might have dispersed or converted in to amorphous form during preparation of microsponges.

j) X-Ray diffraction (X-RD) studies: The X-ray diffraction (X-RD) pattern of drug loaded microsponges showed crystalline nature of microsponges. The resultof X-RD study are shown in Figure No.19. X–ray diffractogram for Itraconazole has shown intense peak between 10 to 20°(2θ) due to its crystalline nature. XRD pattern of drug loaded microsponges showed characteristics peak but at less intensity to pure drug.

k) Stability study: The stability studies were carried out for Itraconazole loaded microsponges at elevated temperature 40°C and 75% RH for 45 days on selected formulation batches (F4 to F9) and. Microsponges were observed for any change in entrapment efficacy. The microsponges did not show any significant change in entrapment efficacyat the end of 45 days. The stability result indicated that all formulations were stable for 45 days. The stability studies were carried out for Itraconazole loaded microsponges at elevated temperature 40°C and 75% RH for 45 days on selected formulation batches (F4 to F9)

4. Summary & Conclusion

a) **Effect of PVA concentration:** In order to know the optimum concentration of surfactant required for formation of microsponges, different concentration of PVA was employed (0.5%, 0.75%, 1%w/v of external phase). The suitable concentration of PVA was found to be 0.5% w/v.
b) **Effect of external phase volume:** To know the effect of external phase volume, different volume of external phase (50,100,200ml) were evaluated for trial batches. The suitable external phase volume was found to be100 ml.
c) **Effect of internal phase volume:** To evaluate the effect of volume of internal phase different volume of methanol and dichloromethane (1:1) ratio and volume 5, 10, 15, 20ml were evaluated. The suitable internal phase volume was found to be 20ml.
d) **Effect of stirring speed on formation of microsponges:** To evaluate the effect of stirring speed on the formation of microsponges, were prepared with different rpm of 300, 500, 800, 1000,1500,2000 keeping all the other variable constant and formed microsponges were evaluated for their drug content and particle size. The suitable stirring speed on the formulation of microsponges was found to be 1000 rpm.
e) **Effect of stirring time on formation of microsponges:** To evaluate the effect of stirring time (duration of stirring) on the formation of microsponges, the different

time interval was employed. The microsponges were prepared by stirring the emulsion for a period of 1hr, 2hrs, 3hrs and 4hrs. The suitable stirring time was found to be 3 hrs.

f) Effect of drug: polymer ratio: In order to evaluate effect of drug to polymer ratio on formulation properties, various ratios of Itraconazole : ethyl cellulose were tried (1:1, 1:2, 1:3, 1:4, 1:5, 1:6, 1:7, 1:8 and 1:9) during trials drug and polymer in ratio 1:4 was found satisfactory to obtained satisfactory yield of microsponges having satisfactory characteristics.

References

[1] Pradhan SK. Microsponges as a versatile tool for drug delivery system: A review. Int J Res Pharm Chem. 2011; 1(2):243-258.
[2] Yerram C, Shaik F, Rubia Y. Microsponges: A novel drug delivery system for controlled delivery of topical drugs. Int J Pharm Res. 2012; 2(2):79-86.
[3] Joshi G, Kaur R. Microsponge anovel drug delivery system, Int Res J Pharm. Biosci.2016; 3(1):01-11.
[4] Patil R, Kemkar V. Microsponge drug delivery: A novel dosage form, Am JPharmTech Res.2012; 2(4):228-251.
[5] Kumar S, Tyagi L. Microsponge drug delivery system MDS: A unique technology for drug delivery of active ingredients, Int J Pharm Sci. Res,2011; 2(12):3069-3080.
[6] Yadav V, Jadav P. Formulation and evaluation of Microsponge gel for topical delivery of antifungal drug, Int J applied Pharm.2017; 9(4):31-37
[7] Tile M K, Pawar Y A. Microsponge a novel strategy for drug delivery, Int J Pure App. Bio Sci. 2015; 3(1):224-235.
[8] Pentewar R S, Kazi S, Bharati R. MDS Technology an approach for topical oral controlled and cosmetic formulations. Res J Pharm BioChem Sci. 2014; 5(3)1170-1190.
[9] Kumari P, Mishra SK. A comprehensive review on novel Microsponges drug delivery approach, Asian J Pharm Clin Res 2016; 2(1):25-30.
[10] 10)Pawar AP, Aditya P. Formulation and evaluation of optimized oxybenzone Microsponge gel for Topical Delivery.2015:1-9
[11] Sharma A, Hoonda A. Formulation and evaluation of topical Microsponges of sertaconazole, World Journal of Pharm Res.2016; 05(11):1444-1461.
[12] Othman MH, Zayed GM. Preparation and evaluation of 5-Flurouracil loaded Microsponge for treatment of colon cancer. J Cancer Sci. 2017:9:307-313.
[13] Tripathi KD. Essentials of medical pharmacology.Jaypee. New Delhi. 2007; 6:254-257.
[14] Rekha L and Manjula BP. Formulation &evaluation of microsponges for topical drug delivery of MometasoneFuroate. Int J Pharm and Sci.2011; 3.
[15] Mahajan AG, Jagtap L. Formulation and Evaluation of microsponge Drug Delivery System, Int Res. J pharm. 2011; 2(10)64-69.
[16] Anju VS,Kuriachan MA. Formulation and evaluation of Terbinafine hydrochloride loaded microsponge based gel for topical sustained delivery. Int J Pharm and p'ceutical Res.2017; 10(2):70-93.
[17] Kumari P, Misra SK. Formulation and evaluation of Tolnaftate microsponges loaded gels for treatment of dermatophytosis, Eur J Pharm Med Res.2017; 4(6): 326-335.
[18] Devasthu M, Pathuri R. Development characterization of Valsartan microsponges by Quasi emulsion technique and development of stirring rate on microsponge formation J. of appl Pharm Sci. 2017 7(1)193-198.
[19] Patel EK, Oswal RJ. Nanosponge and microsponges: a novel drug delivery system. Int J Res Pharm Chem. 2012; 2(2):237-243.
[20] Sinkar NB, Gondkar SB, Saudagar RB. Microsponge a innovative strategy for drug delivery system, current status and future prospects-A Review. Int J Int Pharm Life Sci. 2015; 5(3):226-242.
[21] Shalin KP, Patel DP. Development and validation of UV Spectrophotometric method for estimation of Itraconazole bulk drug and pharmaceutical formulation, Int J of drug dev and res.2011; 3(2):324-328.
[22] Chand SK, Kumar S. Design development and evaluation of Itraconazole loaded transfersomal gel, Int J pharm, 2016; 2(4):198-209.
[23] Kumar CA, Krishna GR. Formulation and characterization of Itraconazole ethmosomal gel for topical application, J Bio. Innov 2017; 6(1):55-64.
[24] Salunkhe PA, Ingale K. UV visible Spectrophotometric method development and validation of Itraconazole in bulk and capsule formulation,Int J Current Res, 2017; 9(1):45436-45439
[25] Rao MK, Rao DN. Method development and validation of Itraconazole by spectrophotometer World J Pharm Res, 2014; 3(10):777-787
[26] Kataria M, Bhandari M. Solubility and dissolution rate enhancement of Itraconazole by solid dispersion technique, Indo American J Pharm Res, 2013; 4(1):849-863.
[27] Battu PK, Tiwari AK.Formulation and evaluation of controlled release drug delivery system, J Pharm Bio Res.2015; 3(2):261-266.
[28] Pathan A, Dhane PR. Develpoment and validation of UV spectrophotometric method for quantitation of Itraconazole form its capsule dosage form, Int J Chem Pharm Res,2017;4(2):2-9.
[29] Bhimavarapu R, Chitra KP. Itraconazole loaded microsponges- A novel carrier system Int J Inv Pharm Sci., 2015; 3(1):953-957.
[30] Pavani V, Vinod M. Design, Formulation and in-vitro evaluation of microsponges based gel for topical delivery of Ketoconazole, Int J. Pharm. Sci. Res.2017;8(10):4222-4229.
[31] Moin A, Tamal K. Fabrication, characterization and evaluation of microsponge delivery system for facillated fungal therapy, J. basic and clini Pharma,2016;7(2):39-47.
[32] Kumar JR, Selvadurai M, Evaluation of antifungal activity of sustained release microsponge enriched Fluconazole gel for Penile Candidiasis in Male Rats, Int. J. Pharm. Tech. Res,2014;6(6):1888-1897.