INTRODUCTION

The female genital tract consists of cervix and vagina. The vagina is extremely flexible, bent canal that is approximately 15 cm long in which posterior wall is larger than frontal wall because of presence of cervical portion of genital tract which is further connected to vestibule. A healthy reproductive woman has an acidic vaginal pH. There is a natural balance of aerobic and anaerobic microbes in vagina. The vagina consists of numerous microbes that exist in equilibrium, and their presence is influenced by various factors such as pregnancy, menstruation, hygiene and infection. Microbes include bacteria, yeast and viruses, in which the vagina is dominated by Lactobacillus species. Lactobacillus helps to prevent such growth of unwanted harmful microbes by various mechanisms such as stimulating the immune system, maintaining the vaginal pH, production of antimicrobial substance such as hydrogen peroxide that acts against pathogens and further competing with other microbes for nutrients. The reduction in lactobacilli results in change in pH of vagina which can lead to other bacterial infections such as development of bacterial vaginosi.

Bacterial vaginosi (BV) occurs as most widely recognized vaginal disorder affecting prolific, premenopausal and pregnant ladies, with a frequency rate extending from 5% to 50%. BV is depicted by an excess of strict or facultative anaerobic microbes and a decrease of lactobacilli especially those involved in hydrogen peroxide production. Female with BV normally account for vaginal inconvenience. Diagnosis test include: The presence of clue cells (20% of cells as clue cells specify the positivity of test). Vaginal discharge (milky white or gray discharge), Whiff test (Fishy odour noted after addition of 10% of potassium hydroxide) OSOM BV Blue test (based on detection of elevated activity of sialidase within the vaginal fluid, blue or green colour indicates that the sample is positive for BV). Its treatment include oral or vaginal antimicrobials such as metronidazole or clindamycin, and...
probiotics. But conventional dosage forms have certain limitations such as lower bioavailability (oral formulations), leakage, and inappropriate drug release, prolong duration of action (vaginal formulations). These limitations are overcome by advanced topical drug delivery system such as vaginal film, which is a type of polymeric drug delivery system that could be applied without using an applicator. Vaginal films are designed to get rapid dispersion or dissolution of API that come in contact with vaginal fluid forming a smooth, viscous, and bio-adhesive gel. As vaginal fluid have some water content so it favours absorption of drugs having certain solubility in water. But for drugs having poor solubility, formulation strategies has to be applied to enhance the solubility. To enhance the solubility of the drug, there are several methods available, such as changing the pH, using co-solvents, using dielectric solvents, using surface active agents, complexation, hydrotropic solubilisation, mixed hydrotropy and mixed solvency. Mixed solvency concept plays an important role to enhance the solubility.

Mixed Solvency Concept

As per the mixed solvency concept proposed by Maheshwari R.K, each and every substance present in the universe has got solubilizing property i.e. all the liquids, gases and solids possess solubilizing power. As per his statement, each substance is solubilizer. A concentrated aqueous solution containing various water soluble substances may act as good solvent for poorly water soluble drugs. By combining various excipients, additive and synergistic solvent actions are expected which has advantage of reducing the toxicities. For a desired solubility enhancement, a single solubilizer may prove toxic for human being but the combination of different excipients in safe smaller concentrations solves the problem of toxicity for same desired solubility of drug.

In present research work mixed solvency concept has been employed to formulate vaginal film of poorly water soluble drug, metronidazole.

2.0 MATERIALS AND METHODS

2.1 Materials: Metronidazole was obtained as a gift sample from Schon pharmaceutical Limited, Indore.

2.2 Estimation of metronidazole:

2.2.1 UV spectrophotometric analysis of metronidazole using 0.1 N HCl

A solution of 10µg/ml of metronidazole in 0.1 N HCl was scanned between 200 to 400 nm on a double beam UV/Visible spectrophotometer (Shimadzu 1700). The UV spectrum of metronidazole drug sample is shown in figure 1.

2.2.2 IR analysis of drug sample:

Metronidazole drug powder was compressed into a pellet along with KBr (KBr pellet technique) using Shimadzu hydraulic press. The FTIR spectrum of drug was recorded in the wave number region of 450-4000 cm⁻¹ on a FTIR spectrophotometer (Shimadzu 8300) and presented in figure 2.

2.2.3 DSC analysis of drug sample:

The DSC study was performed on a Pyris 6 DSC (Jade DSC) differential scanning calorimeter with thermal analyzer. Accurately weighed samples (about 3 mg of samples) were placed in a sealed aluminum pan, before heating under nitrogen flow (20 ml/min) at a scanning rate of 20°C per min from 25 to 350°C. An empty aluminum pan was used as reference. The DSC spectrum of metronidazole drug sample is shown in figure 3.

2.3 Determination of solubility of metronidazole in different solvent medium

Solubility studies in different solvent mediums were carried out by adding an excess amount of drug metronidazole in 5 ml of respective medium and keeping the screw capped tubes containing these solutions, on a mechanical shaker at room temperature for 12 hrs, so that equilibrium solubility can be achieved and solutions were allowed to equilibrate for 24 hrs undisturbed. Then, the solutions were filtered through Whatman grade 41 filter. One ml of the filtrate was suitably diluted with the respective medium. The absorbances of the solutions were measured at 320 nm on a double beam UV/Visible spectrophotometer (Simadzu-1700) against respective reagent blank. The results are recorded in table 2.

2.4 Study of interference of additives in UV spectrophotometric estimation of metronidazole

For determination of interference of additives in the spectrophotometric estimation of metronidazole, the absorbances of solution containing drug (20µg/ml) alone and solution containing drug (20µg/ml) and excipient (2000µg/ml) were recorded against respective reagent blank at 320 nm and results are shown in table 3. A UV-visible recording spectrophotometer (Shimadzu 1700) with 1 cm matched silica cells was employed for spectrophotometric determinations.

2.5 Formulation development of vaginal film:

2.5.1 Solubilizer selection

For the preparation of different blends of solubilizers (% w/v), niacinamide, sodium citrate, caffeine, HP beta cyclodextrin, PVP K 25 (film forming polymer), PEG 4000, propylene glycol, PEG 200, PEG 400, and glycerine were used. Total solute concentration was varied up to 40% w/v concentration and the % solubility of drug (metronidazole) was calculated. To decrease the individual concentration of solublizers as well as individual toxicity of solubilizers, combinations of solubilizers in blend were used to increase the solubility of drug in various ratios. Solubility studies in different aqueous systems of solubilizers were carried out by equilibrium solubility method. The results of solubility studies are shown in table 4.

2.5.2 Polymer selection

For the development of fast dissolving film, HPMC E5, HPMC E50, sodium alginate, PVA, gelatine, CMC were used as film forming polymer (table 5). Blend composition NM:SB:UR:SC =15:10:10:5 (B1) and NM:SB:UR:CF =15:10:10:5 (B2) were selected for film formulation. (NM-Niacinamide, SC-Sodium caprylate, UR-Urea, SB-Sodium benzoate, CF-Caffeine)

Procedure for preparation of single film:

0.8ml blend was taken in a 5ml vial to which accurately weighed 40mg drug was added and covered with the closure then the vial was shaken well to dissolve the drug. Then 0.2 ml plasticizer (glycerine) and 200mg polymer was added to above solution and it was mixed properly using vortex mixer. Then, the film solution was kept undisturbed for 5 hours so as to remove the entrapped air bubble and for proper swelling of the polymer. Then the film solution was poured over a glass petridish and kept for drying at room temperature for 24 hrs.
2.5.2.1 Evaluation of casted polymeric film

a) Folding endurance

Folding endurance was determined by repeatedly folding the film at the same position until it breaks. The number of times the films can be folded without breaking is termed as the folding endurance value.

b) Thickness

The thickness of film was determined by the use of micrometer (Digimatic micrometer, Mitutoyo, Tokyo, Japan) at five locations (centre and four corners) and mean thickness was calculated.

2.8.2.2 Optimization of polymer concentration

Three batches of selected polymer (HPMC E-5) having different concentrations were prepared, casted and their film properties were studied. Results are shown in table 6.

Table 1: Optimized blend for formulation development

| S.No. | Batch code | Blend (8ml) | HPMC E5 (polymer) concentration | Glycerine (plasticizer) concentration | Drug used for 10ml film solution |
|-------|------------|-------------|---------------------------------|--------------------------------------|---------------------------------|
| 1. F1 | B1         | 15%w/v      | 20%w/v                          | 500mg                                 |
| 2. F2 | B1         | 17%w/v      | 20%w/v                          | 500mg                                 |
| 3. F3 | B1         | 20%w/v      | 20%w/v                          | 500mg                                 |
| 4. F4 | B2         | 15%w/v      | 20%w/v                          | 500mg                                 |
| 5. F5 | B2         | 17%w/v      | 20%w/v                          | 500mg                                 |
| 6. F6 | B2         | 20%w/v      | 20%w/v                          | 500mg                                 |

2.5.3 Selection of plasticizer

Five batches of HPMC E-5 with different plasticizers (table 7) having concentration 20% v/v were prepared and casted, after proper drying the films were scrapped out and their film properties were studied.

2.5.3.1 Optimization of plasticizer concentration

Several batches of HPMC E-5 with different concentration range 10%-20% of selected plasticizer glycerine were prepared by adjusting the blend concentration according to the concentration of selected plasticizer so as to get 100% v/v film solution. Then, the prepared batches were evaluated for film properties (table 8).

2.5.4 Optimized batch formula

According to the preliminary studies and studies of selection of excipients, 6 batches were optimized.

2.5.4.1 Method of preparation of fast dissolving film:

500 mg metronidazole was accurately weighed and dissolved in 8ml of respective solubilizer blend taken in a vial of 20ml capacity. Then, 20% v/v glycerine (as plasticizer) and respective concentration (15% or 17% or 20%w/v) of HPMC E-5 (as film forming polymer) were added and properly mixed using vortex mixer. The preparation was placed undisturbed for 5 hrs (for removal of entrapped air bubble and proper swelling of polymer) and then 5ml of prepared film solution was pipetted out and it was uniformly spread over glass petridishes. Then, the film at the same position until it breaks. The number of times the films can be folded without breaking is termed as the folding endurance value.

2.5.4.2 Evaluation parameters:

a) In-vitro release studies

Release study of metronidazole film containing 40 mg drug was performed by setting an assembly in which the film was placed in the beaker containing 30 ml of simulated vaginal fluid on a magnetic stirrer at 50 rpm. Five ml of dissolution media was withdrawn at regular intervals and fresh media was replaced immediately after withdrawal of sample. The sample was filtered and suitably diluted with demineralised water and then analysed in U.V. at 320nm and absorbance was noted against reagent blank (using placebo). Drug release is graphically represented in figure 4.

b) Surface pH

Film was taken and placed in a glass petridish containing 5 ml of simulated vaginal fluid. After wetting of the film, the surface pH of the film was checked by using pH electrode.

c) Disintegration time

10 ml of demineralised water was placed in a beaker and one film was added on the surface of the water and it was shaken briskly. The time was measured until the film was dissolved completely.

d) Thin layer chromatography (TLC) analysis

TLC analysis was done to check any drug-solubilizer interaction (table 10). Acetone was used as solvent for preparation of standard solution of drug and test solution was prepared in aqueous blend of solubilizers. The mobile phase consisted of a mixture of 80 volumes of chloroform, 10 volumes of diethylamine, 10 volumes of ethanol, and 1 volume of water.

e) Stability study of optimized batches F1 and F4:

Stability study of optimized film formulation was carried out for 10 weeks at 2-8°C (refrigerator) and 25°C±2°C (room temperature).
3.1.3 DSC analysis of drug sample: The DSC spectrum of metronidazole was same as reported in literature.
3.2 Determination of solubility of metronidazole in different solvent medium:

Table 2: Solubility data of metronidazole in different solvent medium

| S. No. | Solvent systems          | Solubility (mg/ml) | Inference      |
|--------|--------------------------|--------------------|----------------|
| 1.     | Demineralized water      | 9.8                | Slightly soluble |
| 2.     | Citrate buffer (pH 4.4)  | 9.3                | Slightly soluble |

3.3 Study of interference of additives in UV spectrophotometric estimation of metronidazole: Observing the results from table 3 of drug-excipients interference study, it was concluded that there is nearly no interference in UV spectrophotometric analysis of metronidazole due to excipients.

Table 3: Drug-excipient interference study

| Drug         | Excipient          | Drug conc. (µg/ml) | Solubilizer conc. (µg/ml) | Lambda max (nm) | Absorbance |
|--------------|--------------------|--------------------|----------------------------|-----------------|------------|
| Metronidazole| -                  | 20                 | -                          | 320             | 1.037      |
| Metronidazole| Sodium benzoate    | 20                 | 2000                       | 320             | 1.045      |
| Metronidazole| Niacinamide        | 20                 | 2000                       | 320             | 1.041      |
| Metronidazole| Urea               | 20                 | 2000                       | 320             | 1.043      |
| Metronidazole| Sodium caprylate   | 20                 | 2000                       | 320             | 1.040      |
| Metronidazole| Caffeine           | 20                 | 2000                       | 320             | 1.038      |

3.4 Formulation development of vaginal film:

3.4.1 Solubilizer selection

Table 4: Solubility of drug in aqueous solutions containing various solubilizers

| S.No. | Aqueous blend of solubilizers (%)w/v | Concentration (µg/ml) | Solubility (mg/ml) | Solubility enhancement ratio |
|-------|--------------------------------------|------------------------|--------------------|-------------------------------|
| 1     | DM water                             | 9.8                    | 0.98               | ---                           |
| 2     | PG                                   | 12.47                  | 1.25               | 1.27                          |
| 3     | Glycerine                            | 11.82                  | 1.18               | 1.20                          |
| 4     | PEG 200                              | 10.00                  | 1.00               | 1.02                          |
| 5     | PEG 400                              | 10.12                  | 1.01               | 1.03                          |
| 6     | PVP K 25                             | 11.47                  | 1.15               | 1.17                          |
| 7     | Urea                                 | 17.61                  | 1.76               | 1.79                          |
| 8     | Sodium benzoate                      | 27.94                  | 2.79               | 2.48                          |
| 9     | Niacinamide                          | 30.88                  | 3.09               | 3.15                          |
| 10    | Sodium caprylate                     | 15.14                  | 1.51               | 1.54                          |
| 11    | PG:NM                                | 34.07                  | 3.41               | 3.47                          |
| 12    | PG:SB                                | 26.19                  | 2.62               | 2.67                          |
| 13    | PG:SC                                | 19.11                  | 1.91               | 1.94                          |
| 14    | GLY:NM                               | 31.09                  | 3.11               | 3.17                          |
| 15    | GLY:SC                               | 16.66                  | 1.67               | 1.70                          |
| 16    | GLY:SB                               | 25.94                  | 2.59               | 2.64                          |
| 17    | NM:SB                                | 53.64                  | 5.36               | 5.46                          |
| 18    | UR:SB                                | 48.15                  | 4.82               | 4.91                          |
| 19    | UR:NM                                | 44.62                  | 4.46               | 4.55                          |
| 20    | NM:SB:UR                             | 36.74                  | 3.67               | 3.74                          |
| 21    | SB:SC:NM                             | 27.49                  | 2.75               | 2.80                          |
| 22    | NM:SB:SC                             | 37.96                  | 3.80               | 3.87                          |
| 23    | NM:SB:UR:PEG 4000                    | 41.01                  | 4.10               | 4.18                          |
| 24    | NM:SB:UR:HBC                        | 39.25                  | 3.93               | 4.01                          |
| 25    | NM:SB:UR:CF                         | 52.38                  | 5.24               | 5.34                          |
| 26    | NM:SB:UR:PVP K25                    | 40.30                  | 4.03               | 4.11                          |
| 27    | NM:SB:UR:SC                         | 50.94                  | 5.09               | 5.19                          |

NM-Niacinamide, SC-Sodium caprylate, UR-Urea, SB-Sodium benzoate, CF-Caffeine, HBC-Hβ cyclodextrin.

3.4.2 Polymer selection: HPMC E-5 was taken in further studies because it gave better film properties (table 5) compared to other polymers used to prepare the film.
Table 5: Evaluation of film properties

| S.No. | Polymer      | Physical appearance of film | Folding endurance |
|-------|--------------|-----------------------------|-------------------|
|       |              | B1                          | B2                | B1       | B2       |
| 1     | HPMC-E5      | Transparent, flexible       | Hazy, flexible    | 167      | 183      |
| 2     | HPMC-E50     | Transparent                 | Translucent      | 103      | 118      |
| 3     | Sodium alginate | Opaque, Powdered on scraping | Opaque, cracked   | ---      | ---      |
| 4     | PVA          | Transparent                 | opaque            | 55       | 73       |
| 5     | Gelatin      | Yellowish, translucent, cracked | Yellowish, hazy, hard, brittle | ---      | ---      |
| 6     | CMC          | Opaque, hard, non-foldable | Opaque, brittle   | ---      | ---      |

3.4.2.1 Optimization of polymer concentration: On the basis of selection factors (table 6), 20% w/v concentration of HPMC E-5 was selected and was used in further formulation development studies.

Table 6: Properties of different concentrations of HPMC E-5

| S.No | Property of film | Polymer       | Appearance                        | Folding endurance |
|------|-----------------|---------------|-----------------------------------|-------------------|
|      |                 | HPMC E5 (20%) | HPMC E5 (25%)                    | HPMC E5 (30%)     |
|      |                 | B1            | B2                               | B1                | B2       |
| 1.   | Pourability     | Easy          | Easy                             | Difficult         | Non pourable | Non pourable |
| 2.   | Appearance      | Transparent   | Hazy                             | Transparent       | Hazy     |
| 3.   | Thickness       | 0.05 mm       | 0.07 mm                          | 0.09 mm           | 0.10 mm  |

3.4.3 Selection of plasticizer: On the basis of film properties and effect of plasticizer in polymeric film (table 7), glycerine was selected, it shows better results compared to other plasticizers.

Table 7: Effect of different plasticizers in polymeric casted film

| S.No | Plasticizer (20%v/v) | Polymer | Appearance | Folding endurance |
|------|----------------------|---------|------------|-------------------|
|      | Glycerine            | HPMC E5 | Transparent, flexible, easily peelable | 170 182 |
| 2    | Propylene glycol     | HPMC E5 | Translucent, less flexible             | 80  103 |
| 3    | PEG 200              | HPMC E5 | Hazy, less flexible                     | 73  94  |
| 4    | PEG 400              | HPMC E5 | White, non flexible                       | ---  --- |

3.4.3.1 Optimization of plasticizer concentration: On the basis of film properties (table 8), 20% v/v concentration of glycerine was optimized and used in further studies of formulation development.

Table 8: Effect of different concentrations of selected plasticizers in properties of polymeric film

| S.No | Plasticizer concentration (glycerine) | Appearance       | Thickness (mm) | Folding endurance |
|------|--------------------------------------|------------------|----------------|-------------------|
|      |                                      |                  | B1             | B2                | B1       | B2       |
| 1    | 10%v/v                               | Hazy, hard, brittle | 0.04           | 0.03              | ---      | ---      |
| 2    | 15%v/v                               | Translucent, flexible | 0.04           | 0.05              | 70       | 96       |
| 3    | 20%v/v                               | Transparent, highly flexible | 0.05 mm   | 0.07 mm           | 175      | 186      |

3.5 Evaluations of optimized batches – On the basis of evaluation parameters (table 9) of different batches of films, batch F1 and batch F4 were found to be more appropriate and gave better results as compared to other batches.

Table 9: Evaluation of formulated film batches

| S.No | Batch | Thickness (mm) (average of 5 different positions) | Folding endurance (number) | pH     | Disintegration time (sec) |
|------|-------|---------------------------------------------------|----------------------------|--------|--------------------------|
| 1    | F1    | 0.06                                              | 173                        | 4.67   | 57                       |
| 2    | F2    | 0.08                                              | 103                        | 4.73   | 70                       |
| 3    | F3    | 0.12                                              | 68                         | 4.80   | 82                       |
| 4    | F4    | 0.08                                              | 186                        | 4.69   | 60                       |
| 5    | F5    | 0.09                                              | 125                        | 4.76   | 76                       |
| 6    | F6    | 0.11                                              | 34                         | 4.82   | 89                       |
3.5.1 Thin layer chromatography (TLC) analysis:

Table 10: TLC analysis of pure metronidazole and in blend of solubilizers

| S.No. | Mobile phase                                             | Rf value | Inference                                      |
|-------|----------------------------------------------------------|----------|------------------------------------------------|
|       | Standard solution                                        | B1       | B2                                             |
| 1.    | Chloroform:diethylamine:ethanol:water = 80:10:10:1      | 0.921    | 0.923 0.921                                   |
|       |                                                          |          | No significant change in Rf value hence no interaction between drug and solubilizers |

3.5.2 In-vitro release studies: The % cumulative drug release of optimized batches F1 and F4 in simulated vaginal fluid in 60 minutes were found to be 99.82% and 99.77%, respectively (table 11).

Table 11: In-vitro drug dissolution profile of fast dissolving metronidazole film in simulated vaginal fluid.

| S.No. | Time (min.) | % Cumulative drug release |
|-------|-------------|---------------------------|
|       |             | F1 | F2 | F3 | F4 | F5 | F6 |
| 1.    | 2           | 46.73 | 23.78 | 21.40 | 41.71 | 34.28 | 30.46 |
| 2.    | 5           | 67.79 | 44.63 | 34.81 | 47.81 | 48.82 | 49.60 |
| 3.    | 10          | 83.81 | 62.28 | 47.05 | 66.76 | 73.81 | 63.56 |
| 4.    | 15          | 94.36 | 69.27 | 52.03 | 81.55 | 85.81 | 91.91 |
| 5.    | 30          | 98.72 | 79.67 | 65.36 | 86.64 | 93.47 | 91.68 |
| 6.    | 45          | 99.61 | 91.68 | 73.71 | 94.77 | 94.56 | 84.60 |
| 7.    | 60          | 99.82 | 96.50 | 91.16 | 99.77 | 88.71 | 84.40 |

Figure 4: Graphical representation of % cumulative drug release in simulated vaginal fluid

3.5.3 Stability study of optimized batches F1 and F4:
The developed film of metronidazole was found to be stable for ten weeks at room temperature as well as refrigerated condition. It was found that 97.54% and 97.58% of drug was remaining after stability study at respective temperatures in batch F1 and 98.53% and 96.57% in batch F4.

4.0 CONCLUSION
In the present study, metronidazole, a slightly soluble drug in water was tried to be solubilized by employing the combination of physiologically compatible solubilizers to endeavour its fast dissolving formulations. Amongst all the batches, F1 and F4 batches of prepared fast dissolving films showed better in-vitro dissolution profile, disintegration time and were selected for stability studies. It was found to be stable for 10 weeks. The loading of drug got increased and hence the area of film was reduced. From all the above studies, it was concluded that the approach of mixed solvency concept is novel, safe, cost-effective and user friendly. It also eliminates the problem of toxicity associated with high concentration of water-soluble solubilizers. So, it may be employed in dosage form development of drugs where fast onset of action is required.
REFERENCES

1. Monteiro MR et al, Vaginal films for drug delivery; 2013:102.
2. Mastromarino P et al, Bacterial vaginosis: a review on clinical trials with probiotics. New Microbial, 2013; 36(3):229-38.
3. Eisehnhoch TA et al. Diagnosis and clinical manifestations of bacterial vaginosis. Journal of Obstetrics and Gynaecology, 1988; 158(4):819-28.
4. Dharmik PG et al, Susceptibility pattern of various azoles against candida species causing vulvovaginal candidiasis. Journal of Obstetrics and Gynaecology of India, 2013; 63(2): 135.
5. Ness RB et al, Bacterial vaginosis and risk of pelvic inflammatory disease. Obstet Gynecol, 2004; 104(4):761-769.
6. Johnson TA et al, The effect of pH on release of PGE2 from vaginal and endocervical preparations for induction of labour: an in-vitro study. British Journal of Obstetrics and Gynaecology, 1992; 99(11):877-880.
7. Borumand M et al, Development and in vitro evaluation of a novel contraceptive vaginal mucoadhesive propanolol hydrochloride film. Journal of drug delivery and technology, 2014; 24(6):637-644.
8. Bassi P et al, Polymeric films as a promising carrier for bioadhesive drug delivery: development, characterization and optimization. Saudi Pharmaceutical Journal, 2017; 25(1):32-43.
9. Sudendra RB, Development and characterization of bioadhesive vaginal films of clotrimazole for vaginal candidiasis. Vol. 52. 2010.
10. Dobariar NB et al, A novel iraconazole bioadhesive film for vaginal delivery: design, optimization, and physicochemical characterization. AAPS PharmSciTech, 2009; 10(3):951.
11. Ibrahim A et al, Development and characterization of thermosensitive pluronic-based metronidazole in situ gelling formulation for vaginal application. Acta Pharm, 2012; 62:59-70.
12. Gupta H et al, Ion activated bioadhesive in situ gel of clindamycin for vaginal application. International journal of drug delivery, 2009; 1:32-40.
13. Zhang W et al, Development of a vaginal delivery film containing Efda, a novel anti-HIV nucleoside reverse transcriptase inhibitor. Int. J Pharm, 2014; 461(0): 203-213.
14. Maheshwari RK, "Mixed-Solvency" – A novel concept for solubilization of poorly water-soluble drugs. Journal of Technology and Engineering Sciences, 2009; 1 (1):39-44.
15. Maheshwari RK Solubilization of Ibuprofen by Mixed-Solvency Approach, The Indian Pharmacist, 2009; 8 (87):81-84.
16. Maheshwari RK, Potentiation of solvent character by mixed-solvency concept: A novel concept of solubilization. Journal of Pharmacy Research, 2013; 3 [2]:411-413.
17. Maheshwari RK, Upadhyay N, Jain J, Patani M, Mathuria KC, New spectrophotometric estimation of naproxen tablets formulations employing mixed solvency concept (at 331nm), International Journal of Pharmacy & Technology, 2011; 3 (4):3618-3621.
18. Maheshwari RK, Fouzdar A, "Solid as solvent"- Novel spectrophotometric analytical technique for ornidazole tablets using solids (eutectic liquid of phenol and niacinamide) as solubilizing agents (mixed solvency concept), Indian Drugs, 2015; 52(06): 42-45.
19. Maheshwari RK, Singh S, George P, Fouzdar A, "Solid as solvent"- novel spectrophotometric analytical technique for satranidazole tablets using solids (eutectic liquid of phenol and niacinamide) as solubilizing agents (mixed solvency concept), International Journal of Innovative Research in Pharmaceutical Sciences, 2015; 1(1): 26-29.
20. Jain DK, Patel VK, Bajaj S, Jain N, Maheshwari RK, Novel approach for spectrophotometric estimation of solid dosage forms of tinidazole using solids (eutectic liquid of phenol and niacinamide hydrochloride) as solubilizing agent (mixed solvency concept), World Journal of Pharmacy and Pharmaceutical Sciences, 2015; 4(04):673-769.
21. Jain S et al, Simultaneous estimation of norfloxacin and tinidazole in solid dosage form by uv-spectrophotometry using mixed solvency concept, World Journal of Pharmaceutical and Medical Research, 2018, 4(1):112-117.
22. Maheshwari RK et al; "Solid as solvent"- Novel spectrophotometric analytical method for quantitative estimation of gatifloxacin tablets using solids (eutectic liquid of phenol and lignocaine hydrochloride) as solubilizing agents (mixed solvency concept), European Journal of Biomedical and Pharmaceutical Sciences, 2017; 4(8):644-648.
23. Maheshwari RK, Dahima R, "Solid as solvent"- Novel spectrophotometric analytical technique for quantitative estimation of tinidazole in tablets using solids (eutectic liquid of phenol and lignocaine hydrochloride) as solubilizing agents (mixed solvency concept), Journal of Drug Delivery and Therapeutics, 2017; 7(3):127-130.
24. Rajagopal R, Formulation and evaluation of tinidazole syrup made by mixed solvency concept, Scholars Research Library, 2012; 4(1):170-174.
25. Jain R, Maheshwari RK, George P, Formulation development and evaluation of controlled release tablets of lamotrigine using mixed solvency concept, Bulletin of Pharmaceutical Research, 2015; 5(1):9-14.
26. Maheshwari RK, Gupta P, Gupta H, Formulation development of a model dry injection for reconstitution of poorly water soluble drug ornidazole using mixed solvency concept and its evaluation, International Journal of Science and Research, 2018; 7(4):408-414.
27. Padiyar A, Maheshwari RK; Novel dry injection for reconstitution of aspirin using solid solubilizers, Journal of Drug Delivery and Therapeutics, 2017; 7(7):44-45. https://doi.org/10.10.22270/jddt.v7i7.15822.
28. Patel SK, Maheshwari RK, Formulation development and evaluation of SEDDS of poorly soluble drug made by novel application of mixed-solvency concept, International Journal of Pharmaceutical Research, 2012; 4:51-56.
29. Shilpkar R, Maheshwari RK, Formulation development and evaluation of injection of poorly water soluble drug using mixed solvency concept, International Journal of Pharma and Bio Sciences, 2012; 3:179-189.
30. Solanki SS, Soni LK, Maheshwari RK, Study on mixed solvency concept in formulation development of aqueous injection of poorly water soluble drug, Journal of Pharmaceutics, 2013; 4(2):58-61.