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Original Article

Effects of water-soluble organic co-solvents on the metabolic kinetics of drugs in rat liver microsomes

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Abstract: The objective of this study was to enhance the solubility and dissolution rate of Edoxaban, a drug known for its poor solubility. This was achieved through the utilization of liquid-solid compacts and melt granulation techniques. The solubility assessments were conducted in non-liquid media, followed by thorough evaluations of the resulting formulations. These evaluations encompassed investigations into drug-excipient interactions, flow characteristics, and tablet quality control, employing techniques such as FTIR spectroscopy, DSC, and in vitro dissolution studies. Furthermore, the stability of the formulations was assessed over a three-month period under conditions of 40°C and 75% RH. The outcomes of this study could potentially provide valuable insights into enhancing the therapeutic efficacy of Edoxaban.The study revealed that Maisine CC and Tween 80 exhibited favorable solubility with Edoxaban. Notably, the FT-IR spectra of the drug and polymer did not exhibit any shifts in the major peaks. The tablets formulated in this study were found to be in compliance with Indian pharmacopeial standards and displayed disintegration times ranging from 73 to 85 seconds. Among the various formulations, LSF5 and MGF4 proved to be the most effective. Importantly, even after three months of storage, no significant changes were observed in the formulations, indicating their stability.

Keywords: Edoxaban, Maisine CC, Avicel pH, Aerosil, Gelucire 48/16 pellets, Liquid solid system, melt granulation.

1. INTRODUCTION:

Enhancing the solubility and dissolution rates of poorly water-soluble drugs presents a significant challenge in pharmaceutical research (Yalkowsky and Rubino, 1985; P.K. Lakshmi et al., 2011). These limitations often hinder the oral absorption of drugs (Youn et al., 2006; Sugawara et al., 2005), leading to formulation complexities due to restricted dissolution and permeability (Zhiguo Ma et al., 2018).

Numerous techniques have been explored to improve pharmaceutical solubility, including micronization, nanocrystallization, cyclodextrin inclusion, cocrystallization, solid dispersion, the liquisolid method, and nanoparticle encapsulation. Among these, the liquisolid technique has emerged as a promising approach for enhancing dissolution. It involves converting liquid drugs, suspensions, or solutions into free-flowing, compressible powder

mixtures using carriers and high adsorption capacity coating materials. This method employs biologically safe, non-volatile solvents as liquid vehicles and cellulose, lactose, starch, and their various grades as carriers. In liquisolid compacts, solid drug particles are partially dispersed, resulting in increased aqueous solubility and wetting properties, ultimately leading to improved dissolution rates.

The melt granulation technique is another process used to efficiently agglomerate pharmaceutical powders, utilizing meltable binders without the need for water or organic solvents. Carrier materials and vehicles such as Maisine CC, Avicel pH 102, Aerosil, Tween 80, Gelucire 48/16 pellets, Polyox WSR N-80, and sodium starch glycolate are employed in this technique.

2. MATERIALS AND METHODS

2.1. MATERIALS

Edoxaban was sourced from Aurobindo Pharma Pvt Ltd in Hyderabad.

Gift samples of Maisine CC and Brig 35 were provided by Dr. Reddy's Pvt. Ltd.

Aerosil and Avicel pH 102 were obtained from Sigma-Aldrich in Germany.

Tween 80 was purchased from Sigma Aldrich in Bangalore, India.

Span 80, PEG 400, and PEG 600 were acquired from Sysco Research Laboratories Pvt. Ltd. in Mumbai.

2.2. METHODOLOGY

2.2.1. PREPARATION OF EDOXABAN STANDARD GRAPH

For this experiment, 50 mg of Edoxaban was mixed with 50 ml of citrate/phosphate buffer pH 6.0 in a volumetric flask, creating a concentration of 1000 g/ml. Further dilutions were performed to prepare solutions of lower concentrations. The absorbance of these solutions was measured using a dual-beam UV spectrophotometer to analyze the blank sample. A 10 µg/ml standard solution of Edoxaban in Citrate/Phosphate Buffer pH 6.0 was scanned on the UV spectrophotometer to determine the λ max of Edoxaban. A standard graph was plotted using the absorbance values at different concentrations, and the correlation coefficient (R2) was calculated. This experiment aimed to generate essential data for future research and experiments in this field.

2.2.2. SOLUBILITY STUDIES

The solubility of Edoxaban in the non-volatile liquid vehicles used to create liquisolid systems was investigated by creating saturated solutions of the drug in these vehicles. The drug content of these solutions was spectrophotometrically analyzed. To create systems with an excess of the drug, Edoxaban was combined with specific quantities of each solvent in 7ml screw-capped vials. These mixtures were agitated for 24 hours on an automated test tube shaker and allowed to settle for an additional 2 hours. The vials were then centrifuged at 2500 rpm to settle any undissolved crystalline material and produce a clear supernatant. Precisely measured portions of the filtered residual solutions were diluted with methanol, and their drug concentrations were determined spectrophotometrically at 296 nm.

2.2.3. FLOWABLE LIQUID RETENTION POTENTIAL

The suitability of a liquisolid system for achieving adequate flow rate and compressibility (R) depends on the liquid load factor (Lf) and the excipient ratio. These parameters are connected as follows:

$$Lf = \Phi CA + \Phi Co\ (1/R)$$

2.2.4. PREPARATION OF LIQUISOLID COMPACTS FOR EDOXABAN

Measured amounts of the drug and solvent were mixed in a mortar, along with the carrier and coating material. Sodium starch glycolate was added to the previously mentioned mixture to ensure homogeneity. The resulting blend was shaped into plugs and filled into capsules as specified in Table 1.

Table1:FormulationofLiquisolidSystemsofEdoxaban

Formulations	**LSF1**	**LSF2**	**LSF3**	**LSF4**	**LSF5**	**LSF6**	**LSF7**	**LSF8**
Drug: Liquid	1:2	1:4	1:2	1:4	1:2	1:4	1:2	1:4
Ca:Co(R)	20	20	40	40	20	20	40	40
Lf	0.28	0.28	0.198	0.198	0.351	0.351	0.261	0.261
Drug(mg)	30	30	30	30	30	30	30	30
Maisine CC(mg)	60	120	60	120	-	-	-	-
Tween 80 (mg)	-	-	-	-	60	120	60	120
Avicel pH 102(mg) Q	375	624.9	545.2	757.4	908.88	512.16	413.20	688.70
Aerosil(mg) q	18.74	31.248	13.63	22.72	15.36	25.63	10.32	17.21
SSG (mg) 5%	24.16	40.31	32.44	54.07	20.63	34.368	25.65	42.79
Capsulesize '0'	163	163	163	163	163	163	163	163
Unit weight of blend (mg)	**507.916**	**846.518**	**681.36**	**1135.6**	**433.370**	**722.136**	**539.193**	**898.716**
Total weight of filled capsule(mg)	**703.516**	**1042.11**	**876.96**	**1331.27**	**628.970**	**717.736**	**734.79**	**1094.31**

R =Carrier:Coating(Q:q)-[MicrocrystallineCellulose:Aerosil)

LiquidloadFactor:Lf=W/Q

LV: Liquid Vehicle(MaisineCC&Tween80)

2.2.6. MELT GRANULATION TECHNIQUE FOR EDOXABAN:

Procedure: The polymer was precisely weighed and then transferred into a porcelain dish. This dish was placed onto a hot plate and heated to 55ºC for Gelucire 48/16 pellets and 65ºC for Polyox WSR N-80 until the polymer completely melted. Subsequently, the porcelain dish was removed from the hot plate, and a measured quantity of the drug was added. The mixture was thoroughly stirred to achieve a uniform blend. After the mixture solidified, it was broken into smaller pieces and sifted through a #40 mesh sieve. Avicel pH 102 and SSG were also sifted through a #40 mesh sieve and then added to the blend,

where they were thoroughly mixed. Additionally, Aerosil and Magnesium stearate were sifted through a #60 mesh sieve and blended with the rest of the mixture. Finally, this resulting blend was filled into the capsules as specified in Table 2.

Table2:FormulationofEdoxabanbyMelt Granulation Technique

Formulations	**MGF1**	**MGF2**	**MGF3**	**MGF4**	**MGF5**	**MGF6**	**MGF7**	**MGF8**
Drug:Polymer	1:0.25	1:0.5	1:1	1:2	1:0.25	1:0.5	1:1	1:2
Drug(mg)	30	30	30	30	30	30	30	30
Gelucire 48/16 pellets(mg)	7.5	15	30	60	-	-	-	-
PolyoxWSRN-80(mg)	-	-	-	-	7.5	15	30	60
Avicel pH 102 (mg)	18.6	11.1	52.2	22.2	18.6	11.1	52.2	22.2
SSG(mg) 5%	3	3	6	6	3	3	6	6
Aerosil (mg) 0.5%	0.3	0.3	0.6	0.6	0.3	0.3	0.6	0.6
Magnesium stearate (mg) 1%	0.6	0.6	1.2	1.2	0.6	0.6	1.2	1.2
Capsulesize	Size2	Size2	Size2	Size2	Size2	Size2	Size2	Size2
Capsule weight (mg)	63	63	63	63	63	63	63	63
Weightofblend (mg)	60	60	120	120	60	60	120	120
Total weight of filled capsule (mg)	123	123	183	183	123	123	183	183

3.RESULTSAND DISCUSSION:

3.1.SOLUBILITYSTUDIES:

Edoxaban's saturated solution in millilitres per percent with the solvents under examination wascalculated using extrapolation from solubility experiments of Edoxaban in non-volatile liquid vehicles. In Table 3, results are displayed.

Table3: SolubilityStudies

S.No	**Non-Volatile Liquid Vehicles**	**Absorbance at 296.00nm**	**Solubility (µg/mL)at296.00 nm**
1	PEG400	0.051	172.7052
2	PEG600	0.086	293.9383

4	MAISINE CC	0.216	744.2327
6	BRIG 35	0.072	245.4450
7	TWEEN 80	0.194	668.0290
8	SPAN 80	0.120	441.7076

Observation: Fromtheabovedataobtained,MaisineCC(744.2327µg/mL)&Tween 80(668.0290 µg/mL) were foundtohavegoodsolubilitywithEdoxaban.

3.2.FLOWABLELIQUIDRETENTION POTENTIAL:

Table 4 shows the results for the flowable liquid-retention capability of the carrier and coat materials.

Table4:FlowableLiquid Retention Potential

Material	MaisineCC	Tween 80
Avicel pH 102	0.135	0.1842
Aerosil	3.6	3.6

3.3. FTIR ANALYSIS OF MEDICATION AND POLYMER COMPATIBILITY

We utilized FT-IR spectroscopy to investigate potential interactions among the components within the optimal composition. The results revealed that there were no significant changes in the FT-IR spectra of the drug and polymer, either individually or in their physical combination. The absence of substantial alterations in the spectral peaks indicates that the properties of both the drug and polymer in the formulation remained unaltered. This confirmed the compatibility between the drug and polymer.

The IR spectrum of Edoxaban displayed characteristic peaks at 1614.41 cm-1 (C=O stretching), 1503.28 cm-1 (N-H stretching), 1378.41 cm-1 (-CH3), and 683.79 cm-1 (C-H stretching). These characteristic bands' positions remained stable when the drug was combined with individual excipients or when the formulation was derived during the investigation. Consequently, it was established that the drug retained its identity and did not undergo any chemical interactions with the utilized polymers.

3.4. EVALUATION:

All formulations exhibited favorable flow characteristics, as indicated by angle of repose values ≤ 35. Interestingly, formulations with lower Drug:Liquid ratios demonstrated superior flow at specific excipient ratios, possibly attributed to reduced liquid content resulting in weaker cohesive forces. Importantly, all formulations fell within an acceptable range for Carr's index (CI) values, ranging from 11.21 to 15.36, and Hausner ratio values between 1.121 and 1.302, indicating fair to reasonable flow properties. Among all the formulations, LSF8 displayed the lowest CI and Hausner values, likely due to its lower Drug:Liquid ratio and a 20% excipient ratio. Furthermore, all capsules disintegrated within 73 seconds, as detailed in Table 5.

Table5:FlowProperties of FormulationsLSF1-LSF8(Liquid solid Compacts Technique forEdoxaban)

Formulations	LSF1	LSF2	LSF3	LSF4	LSF5	LSF6	LSF7	LSF8
Angleof repose	34	35	32.1	33	31	30	33	29.5

Bulkdensity	0.259	0.268	0.254	0.266	0.251	0.236	0.241	0.230
Tapped density	0.348	0.366	0.301	0.309	0.328	0.315	0.306	0.314
CI	14.99	13.55	15.36	12.98	14.12	11.98	13.36	11.21
Hausnersratio	1.265	1.276	1.286	1.302	1.299	1.289	1.267	1.121
DT (min:sec)	1:07	1:01	0:59	1:05	0:55	1:05	1:04	1:13

3.5. IN VITRO DRUG RELEASE

Upon breaking open the liqui-solid capsule, the suspended particles in the dissolving solvent primarily consisted of drug particles in a state of molecular dispersion. These drug particles in the liqui-solid formulations were dispersed within a carefully selected hydrophilic liquid vehicle. Since the medication was already in a solubilized state, all the formulations exhibited nearly complete drug release within just 15 minutes.

While it's possible that more of the liquid medication may have been delivered in table 6, the immediate release observed in LSF2, LSF5, and LSF8 appeared to be influenced by the higher excipient ratio, leading to greater drug release.

Table6: PercentageCumulativeDrugRelease for theFormulationsLSF1- LSF8

	% Cumulative drugrelease							
Time								
0	0	0	0	0	0	0	0	0
5	34.11	39.44	42.97	49.44	61.77	52.37	53.77	50.17
10	42.19	45.77	58.44	60.17	72.11	62.77	64.77	61.77
15	56.77	59.44	64.77	69.22	80.11	70.11	71.22	73.77
20	62.11	68.11	71.44	79.33	89.77	80.17	81.22	82.77
30	79.22	89.44	91.22	92.77	96.77	93.77	94.77	92.17
45	92.77	96.22	99.79	100.03	100.45	99.89	99.98	100.35

3.6.EVALUATION:

DETERMINATION OFFLOWPROPERTIESAND DISINTEGRATION TIME:

Table7:FlowPropertiesof Formulations MGF1-MGF8(EdoxabanebyMelt Granulation Technique)

Formulations	**MGF1**	**MGF2**	**MGF3**	**MGF4**	**MGF5**	**MGF6**	**MGF 7**	**MGF8**
Angle of repose	30	29	21.6	18	25.3	28	29	23.2

Bulk density	0.495	0.467	0.458	0.358	0.396	0.413	0.425	0.443
Tapped density	0.594	0.574	0.556	0.451	0.498	0.512	0.536	0.541
CI	19.36	21.87	25.6	12.64	18.66	21.67	22.47	23.98
Hausner ratio	1.354	1.199	1.298	1.112	1.254	1.287	1.279	1.654
DT (inminutes)	1:12	0:58	1:12	1:25	1:11	1:01	1:18	1:03

3.7. IN VITRO RELEASE OF DRUGS :

Dissolution testing was conducted on all the formulations using 900 ml of 0.1N HCl at a rotation speed of 50 rpm, resulting in complete drug release. It was observed that as the concentration of polymer increased, there was an improvement in dissolution rates. However, at higher polymer concentrations, the formation of more rigid granules occurred, leading to reduced initial drug release, particularly noticeable in the cases of MGF6 and MGF7.

Comparatively, the Polyox formulations exhibited lower initial drug release in comparison to the Gelucire formulation. This distinction arises from the fact that Polyox is a water-swellable polymer, which hampers drug diffusion, while Gelucire is a water-soluble excipient, facilitating quicker dissolution.

Based on the drug release profiles, the MGF4 formulation demonstrated superior results, as outlined below.

Table8: % CumulativeDrug Releasefor theFormulationsMGF1- MGF8

	%CumulativeDrugRelease							
Time (in minutes)	**MGF1**	**MGF2**	**MGF3**	**MGF4**	**MGF5**	**MGF6**	**MGF7**	**MGF8**
0	0	0	0	0	0	0	0	0
5	30.44	37.99	38.99	49.66	33.49	38.11	40.19	43.66
10	44.11	49.33	52.19	62.11	53.77	59.11	57.44	56.11
15	55.22	59.11	62.44	71.55	67.22	65.44	64.22	60.11
20	65.78	69.77	71.33	85.66	79.44	76.88	79.11	80.11
30	87.17	90.44	91.77	96.77	91.77	86.33	89.55	85.22
45	98.47	99.01	97.44	100.98	100.01	99.66	100.04	99.98

3.8.EDOXABAN STABILITY INFORMATION:

DSCStudies: Differential scanning calorimetry (DSC) serves as a crucial tool for assessing the stability of formulation blends in both the liqui-solid and hot granulation approaches. The selection of LSF5 and MGF4 as the most suitable formulations led to in vitro stability evaluations, the outcomes of which are presented in Table 9. The optimized formulation underwent stability testing over a three-month period at temperatures of 20°C, 40°C, and a relative humidity of 75%.

Table9: Stability data of Edoxaban

		Initial	**After3 months**
Formulation			
Liquid solid Systems	LSF5	100.45	100.99
Melt-Granulation Technique	MGF4	100.98	101.06

4. CONCLUSION:

Edoxaban served as a prototype medication to assess the potential of both the Liquisolid system and the melt-granulation process in enhancing the dissolution characteristics of water-insoluble pharmaceuticals. These two techniques have gained considerable attention and significance in recent times for improving drug solubility.

The Fourier-transform infrared (FT-IR) spectrum of the drug reveals no discernible differences in its properties compared to the polymers incorporated in its formulation. At specific excipient ratios, the drug solution exhibits favorable flow properties. The notably increased surface area of the drug particles achieved through molecular dispersion could account for the observed enhancement in dissolution rates within liquid formulations. Ultimately, MGF4 and SPF5 were identified as the most promising formulations.

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