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FORMULATION AND EVALUATION OF TERNARY SYSTEM FOR SOLUBILITY ENHANCEMENT OF A SECOND GENERATION COX-II INHIBITIOR

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ABSTRACT

Etoricoxib is a highly selective COX-II inhibitor, used to treat pains of different etiologies. Etoricoxib has low aqueous solubility (201µg/ml) and high permeability and therefore classified as BCS class II drug. By formulating these drugs with cyclodextrins as inclusion complexes have shown to increase the bioavailability. Cyclodextrins when used as Complexing agents, enhance the solubility of poor water soluble lipophilic drugs. The objective of the present work is to formulate Etoricoxib cyclodextrin complexes by using ternary systems as Citric acid, Tartaric acid and PVP K-30 in order to enhance solubility and evaluate the enhanced solubility by *in-vitro* dissolution. The formulated complexes were evaluated for drug release by *in-vitro* dissolution. The prepared complexes were confirmed and characterized by FT-IR, DSC, SEM and XRD. Complexation with cyclodextrins is an effective method in order to improve the aqueous solubility of Etoricoxib. The release of Etoricoxib from complexes prepared with Hydroxy propyl β cyclodextrin and citric acid (1:1:2M) by lyophilization method shows a drug release 100.85%, significantly higher than other cyclodextrin ternary complexes.

Keywords: Etoricoxib, cyclodextrins, Kneading method, Solvent evaporation method

INTRODUCTION

Solubility is defined in quantitative terms as concentration of a solute in a saturated solution at a certain temperature and a qualitative way it may be defined as the spontaneous interaction of two or more substances to form a homogenous molecular dispersion[1].

The drug substance administered by any route must possess some aqueous solubility for systemic absorption[2]. Solubility is one of the important parameter to achieve desired concentration of drug in systemic circulation for therapeutic response. If a compound has a low aqueous solubility, it may be subject to dissolution rate limited absorption with the gastro-intestinal (GI) tract residence time. Recently, the importance of solubility in biopharmaceutical terms, has been highlighted by its use in Biopharmaceutics Classification System (BCS) described by Amidon *et al* (1995)[3].

The BCS is a scientific framework for classifying a drug substance based on its aqueous solubility and intestinal permeability. When combined with the in vitro dissolution characteristics of the drug product, the BCS takes into account three major factors: solubility, intestinal permeability, and dissolution rate, all of which govern the rate and extent of oral drug absorption from IR solid oral-dosage forms. It classifies drugs into four classes Solubility

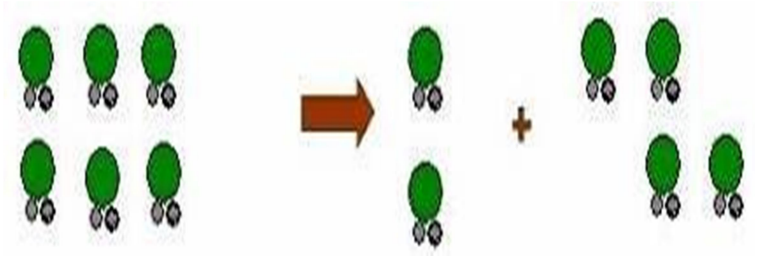
Process of solubilisation[4]:

The process of solubilisation involves the breaking of inter-ionic or intermolecular bonds in the solute, the separation of the molecules of the solvent to provide space in the solvent for the solute, interaction between the solvent and the solute molecule or ion.

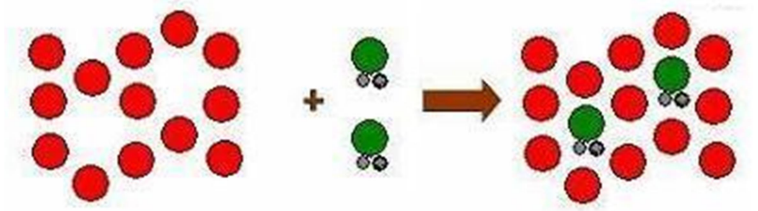
A variety of solubilization techniques have been studied and widely used for enhancement of solubility. Many estimates up to 40% of new chemical entities which are having problem in solubility, discovered by pharmaceutical industry. Today, as per BCS-II, the numbers of drugs are poorly soluble and lipophilic in nature. Aqueous solubility lesser than 1µg/ml will definitely create a bioavailability problem and thereby affects the efficacy of the drug. There are number of methods through which aqueous solubility of the drug can be increased[9].

Step 1: Holes opens in the solvent

Step2: Molecules of the solid breaks away from the bulk



Step 3: The freed solid molecule is intergrated into the hole in the solvent



TECHNIQUES FOR SOLUBILITY ENHANCEMENT[4,5]

1. **Micronization:** Particle size reduction leads to increase in the effective surface area resulting in enhancement of solubility and dissolution velocity of the drug. Particle size reduction methods include re-crystallization of the solute particles from solutions using liquid anti-solvents, along with labor intensive techniques like crushing, milling, grinding, freeze drying and spray-drying.
2. **Use of surfactant:** Surface active agents (surfactants) are substances which at low concentrations, adsorb onto the surfaces or interfaces of a system and alter the surface or interfacial free energy and the surface or interfacial tension. Improvement of drug solubility by using the amphiphilic surfactants is due to lowering of surface tension between drug and solvent, improvement of wetting characteristics and micellar solubilization of the drugs.

3. **Use of co-solvent:** Co-solvent addition is a highly effective technique for enhancement of solubility of poorly soluble drugs. It is well-known that the addition of an organic co-solvent to water can dramatically change the solubility of drugs. Weak electrolytes and non polar molecules have poor water solubility and it can be improved by altering polarity of the solvent. This can be achieved by addition of another solvent. This process is known as co solvency. Solvent used to increase solubility is known as co solvent. The use of mixed solvent system is often necessary in pharmaceuticals when a drug is poorly soluble.
4. Co-solvents such as ethanol, propylene glycol, glycerin, sorbitol and polyoxyethylene glycols, dimethylsulfoxide, ethanol and N, N dimethyl formamide can be used.
5. **Hydrotropy method:** Hydrotropy is a solubilization phenomenon whereby addition of large amount of a second solute results in an increase in the aqueous solubility of another solute. The term "Hydrotropy" has been used to designate the increase in aqueous solubility of various poor water soluble compounds due to the presence of a large amount of additives.
6. **Use of salt forms:** A major improvement in solubility and dissolution rate can be achieved by forming a salt. Salts of acidic and basic drugs have, in general, higher solubilities than their corresponding acid or base forms.
7. **Solvent deposition:** In this technique drug is dissolved in a solvent like methylene chloride to produce a clear solution. The carrier is then dispersed in the solution by stirring and the solvent is removed by evaporation under temperature and pressure. The resultant mass is then dried, pulverized, and passed through a sieve. The increase in the dissolution rate is ascribed to the reduced particle size of the drug deposited on the carrier and enhanced wettability of the particles brought about by the carrier.
8. **Solubilizing agents:** Solubilizing materials like superdisintegrants such as crospovidone, crosscarmellose sodium and sodium starch glycolate used as solubilizing agents in many

formulations which increase the solubility and dissolution rate of poorly water soluble drugs.

9. **Supercritical fluid method:** A supercritical fluid (SCF) can be defined as a dense noncondensable fluid which is a novel nanosizing and solubilisation technology whose application has increased in recent years. A SCF process allows the micronization of drug particles within sub micron levels.
10. Supercritical fluids are fluids whose temperature and pressure are greater than critical temperature (Tc) and critical pressure (Tp). At near-critical temperature, SCFs are highly compressible, allowing moderate changes in pressure to greatly alter the density and mass transport characteristics of a fluid that largely determine its solvent power. Once the drug particles are solubilised within SCF, they may be recrystallized at greatly reduced particle size. Carbon dioxide and water are the most commonly used supercritical fluids. The SCF process can create nanoparticulate suspensions of particles 5–2,000 nm in diameter.
11. **Complexation:** Considerable increase in solubility and dissolution of the drug has been achieved by the use of cyclodextrins. Cyclodextrins are non-reducing, crystalline, water soluble, cyclic, oligosaccharides. Cyclodextrins consist of glucose monomers arranged in a donut shape ring. Three naturally occurring cyclodextrins are α-Cyclodextrin, β-Cyclodextrin, and γ- Cyclodextrin. The complexation with cyclodextrins is used for enhancement of solubility.
12. **Solid dispersion:** Solid dispersion (SD) technique has been widely used to improve the dissolution rate, solubility and oral absorption of poorly water-soluble drugs. Solid dispersion is defined as the dispersion of one or more active ingredients in an inert excipient or matrix (carrier), where the active ingredients could exist in finely crystalline, solubilised or amorphous state.

The enhanced solubility and dissolution rate of drugs from solid dispersions is based on following mechanisms:

1) Reduction in particle size provides large surface area. 2) Particles with improved wettability and dispersibility of drug. 3) Particles with higher porosity. 4). Drugs in

amorphous state. 5) Solubilizing effect on the drug by water soluble carrier. 6) Formation of metastable dispersion.

Various pharmaceutical approaches for the preparation of SDs include co-precipitation, lyophilization, spray drying, melting solvent method, melt extrusion method, solvent evaporation, fusion and powder mixing methods.

Melting and solvent evaporation methods are the two major processes of preparing solid dispersions.

a. Melting method: The melting or fusion method, first proposed by Sekiguchi and Obi involves the preparation of physical mixture of a drug and a water-soluble carrier and heating it directly until it melts. The melted mixture is then solidified rapidly in an ice-bath under vigorous stirring. The final solid mass is crushed, pulverized and sieved. However many substances, either drugs or carriers, may decompose during the fusion process which employs high temperature. It may also cause evaporation of volatile drug or volatile carrier during the fusion process at high temperature.

Some of the means to overcome these problems could be heating the physical mixture in a sealed container or melting it under vacuum or in presence of inert gas like nitrogen to prevent oxidative degradation of drug or carrier.

b. Solvent evaporation method: The solvent evaporation method consists of the dissolving the drug and polymeric carrier in a common solvent, such as ethanol, chloroform, or a mixture of ethanol, dichloromethane, which is evaporated until a clear, solvent free film is left. Normally, the resulting films are pulverized and milled.

In this method, the thermal decomposition of drugs or carrier can be prevented, since organic solvent evaporation occurs at low temperature. The use of organic solvents, the high preparation cost and the difficulties in completely removing the solvent are some of the disadvantages associated with solvent evaporation methods.

Cyclodextrins and derivatives[6-8]

Complexation with cyclodextrin (CD) is known as an effective method for enhancing dissolution properties and bioavailability of poorly soluble drugs. For several reasons, including toxicity, cost and dosage, the amount of CD in the dosage form has to be reduced as far as possible. Different approaches can be considered for achieving this goal. The first is the use of chemically modified CDs, which present a higher solubility in water. The second method consists in adding a water-soluble polymer

like PVP or methylcellulose with the aim to increase the solubility of both the complex and the drug itself. The third method is the use of an acidic ternary compound. In the case of a basic drug, the acidic agent, for instance, citric acid, tartaric acid, promotes the solubilization of the guest molecule both by forming a salt and by increasing the stability constant of the complex[9].

Applications of cyclodextrin[10].

Since each guest molecule is individually surrounded by a cyclodextrin (derivative) the molecule is micro-encapsulated from a microscopical point of view. This can lead to advantageous changes in the chemical and physical properties of the guest molecules.

- Stabilization of light- or oxygen-sensitive substances.
- Modification of the chemical reactivity of guest molecules.
- Fixation of very volatile substances.
- Improvement of solubility of substances.
- Modification of liquid substances to powders.
- Protection against degradation of substances by microorganisms.
- Masking of ill smell and taste.
- Masking pigments or the color of substances.
- Catalytic activity of cyclodextrins with guest molecules.

The Aim and objectives of the project is as follows

- ✓ To increase the solubility of Etoricoxib by preparation and evaluation of cyclodextrin complexation.
- ✓ Evaluation of prepared tablets and their comparison with marketed product.
- ✓ To evaluate pre-compression and post-compression properties of conventional tablet.
- ✓ To evaluate the drug release property of prepared conventional tablets and their comparison with marketed product.

MATERIALS AND EQUIPMENTS[11]

Table 1: List of materials used

	Name	Source
Drug	Etoricoxib	Zydus Cadilla
Cyclodextrins	β- Cyclodextrin	Signet pharma
	Hydroxy propyl β-cyclodextrin	Roquette pharma
Chemicals	Potassium di-hydrogen phosphate	S.D. Fine chemicals
	Sodium hydroxide	
	Methanol	Loba Chemie
	Magnesium stearate	
	PVP K-30	

Ingredients	Citric acid	
	Tartaric acid	
	Microcrystalline cellulose	Supplier
	Talc	
	Croscarmellose sodium	

Table 2: list of equipment used

Equipment name	Manufacturing company
Electronic balance	Shimadzu Corporation, Japan.
pH meter	Digisun Electronics, Mumbai.
Dissolution apparatus	Lab India Disso 2000, Chennai.
UV/VIS Spectrophotometer	Techcomp, UV 2300
Spray dryer	Labultima, Model: LU 222 Advanced
Freeze dryer	Modulyod 230
Compression machine	Rimek, 10 stationary punching machine
Infrared spectrophotometer	Shimadzu Corp, Japan
Differential scanning colorimeter	DSC Q20 V24.2 Build 107
X-Ray diffraction	Philips PW 1710.
Scanning electron microscopy	Model JSM 840A, Jeol, Japan.

METHODOLOGY[12-17]

Formulation Development of Etoricoxib Tablets by Direct Compression method

Direct compression is the preferred method for the preparation of tablets. It offers several advantages. Notable among them are (i) It is economical compared to wet granulation since it requires fewer unit operations (ii) More suitable for moisture and heat sensitive APIs since it eliminates wetting and drying steps (iii) Changes in dissolution profiles are less likely to occur in tablets made by direct compression method on storage than in those made from granulations. This is extremely important because the official compendium now requires dissolution specifications in most solid dosage forms. Disintegration or dissolution is the rate limiting step in absorption in the case of tablets of poorly soluble API prepared by wet granulation. The tablets prepared by direct compression disintegrate into API particles instead of granules that directly come into contact with dissolution fluid and exhibits comparatively faster dissolution.

Table 3: Composition development of F1 batch

INGREDIENTS	F1 (mg/tablet)
ETR- HP β cd- citric acid complexes	310
Croscarmellose sodium	5
Microcrystalline cellulose	183
Magnesium stearate	1
Talc	1
Total weight of tablet	500

RESULTS

Melting point:

Table 4: Melting point of pure drug by Thiel's tube method

No	Reported	Observed		
1.	134°C – 135°C	Trial 1	Trial 2	Trial 3
		136°C	135°C	135°C

Calibration curve:

Table 5: Calibration curve of Etoricoxib in phosphate buffer pH 6.8.

Concentration in µg/ml	Absorbance
6	0.252
8	0.339
10	0.434
12	0.521
14	0.606
16	0.702
18	0.791
20	0.869

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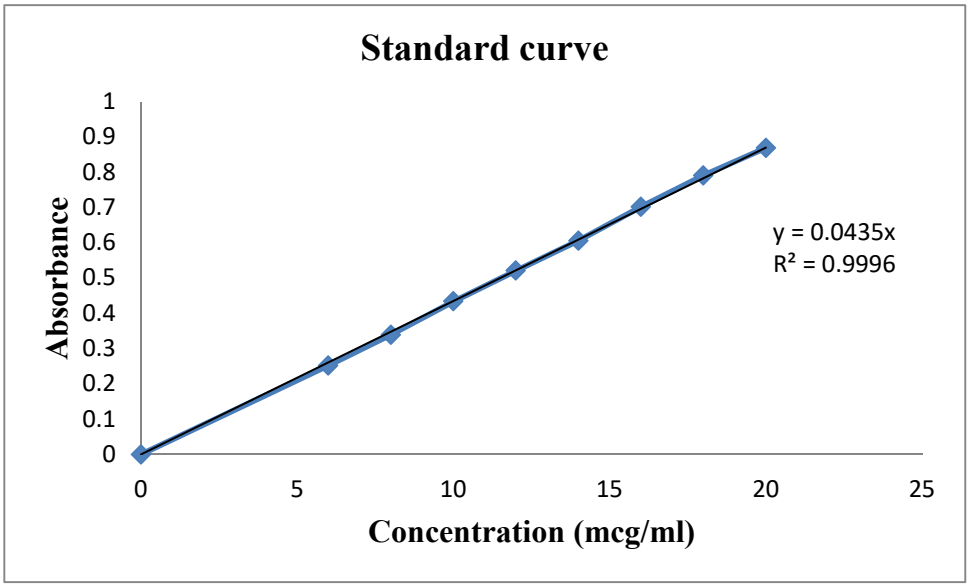

Figure 1: Standard curve of Etoricoxib in phosphate buffer pH 6.8 at 284nm

PHASE SOLUBILITY STUDIES:

Table 6: Phase solubility studies of Etoricoxib: HP β cd: Citric acid (0.2%w/v) complexes

Concentration of HP β-cd in mmol/L with 0.2%w/v citric acid	Concentration of Etoricoxib (mmol/L)
2	25.35
4	29.34
6	32.79
8	36.43
10	39.56
12	38.86

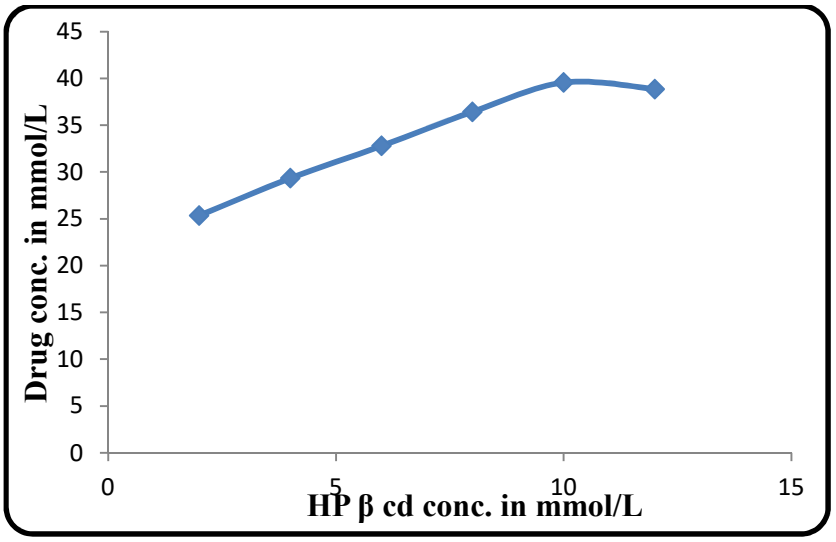

Figure 2: Phase solubility curve of HP β CD–etoricoxib- 0.2% of citric acid in phosphate buffer 6.8pH.

Pre compression parameters

Table 7: Loose bulk density, Tapped bulk density and Carr's compressibility index

Formulations	Loose bulk density (g/ml)	Tapped bulk density (g/ml)	% Compressibility	Hausner's ratio
F1	0.449	0.492	8.739	1.09

Table 8: Evaluation parameters of prepared Etoricoxib Tablets

Formulation	Hardness (Kg) (n=6)	Disintegration (sec)	Thickness (mm) (n=6)	Weight variation (mg) (n=20)	Friability (%)	Drug content (%)
F1	4.4±0.68	560	3.30±0.019	619±2.1	0.59	99.4±0.12

DISSOLUTION CHARACTERISTICS

Table 9: Dissolution studies of Etoricoxib-βcd-Citric acid complexes in phosphate buffer pH 6.8.

Percentage drug release* ± SD				
Time	Kneading method		Solvent evaporation	
	1:1:1M	1:1:2M	1:1:1M	1:1:2M
0	0	0	0	0
5	56.15±0.24	61.24±0.19	58.23±0.34	62.25±0.24
10	62.43±0.79	68.14±0.76	62.54±0.86	66.77±0.44
15	67.23±0.39	73.32±0.51	68.32±0.45	71.26±0.12
30	71.21±0.63	75.12±0.34	72.45±0.31	75.78±0.33
45	74.76±0.23	79.29±0.98	75.13±0.22	79.97±0.62
60	76.24±0.42	83.85±0.32	78.46±0.13	84.81±0.41

*= average cumulative drug release of triplicate samples (n=3).

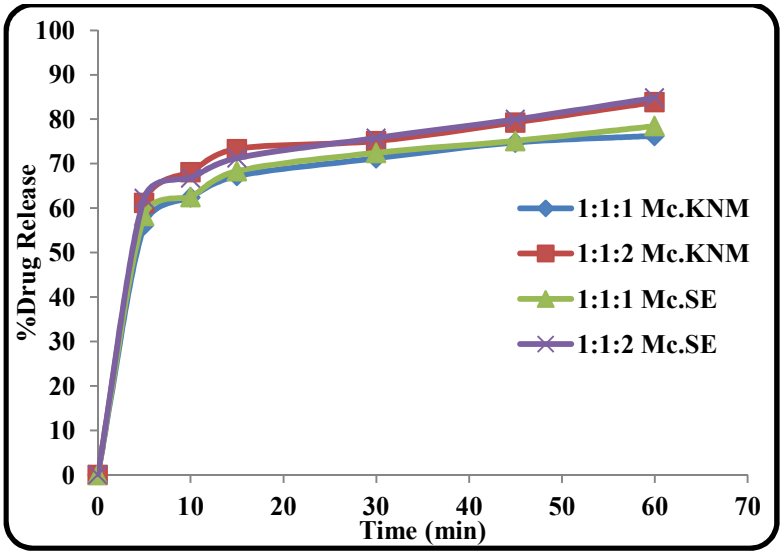

Figure 3: Dissolution profile of Etoricoxib-βcd-Citric acid complexes in phosphate buffer pH 6.8.

Table 10: Dissolution studies of Etoricoxib-HP β cd- Citric acid complexes in phosphate buffer pH 6.8.

Percentage drug release* ± SD				
Time	Kneading method		Solvent evaporation	
	1M	2M	1M	2M
0	0	0	0	0
5	62.56±0.17	64.75±0.12	64.22±0.47	68.23±0.42
10	64.72±0.42	67.62±0.52	67.53±0.57	73.45±0.58
15	68.43±0.95	72.43±0.88	68.31±0.53	79.12±0.67
30	67.42±0.76	76.84±0.75	71.46±0.75	84.34±0.85
45	72.47±0.42	81.74±0.43	75.91±0.94	88.23±0.42
60	74.64±0.56	84.57±0.19	82.68±0.41	91.21±0.48

*= average cumulative drug release of triplicate samples (n=3)

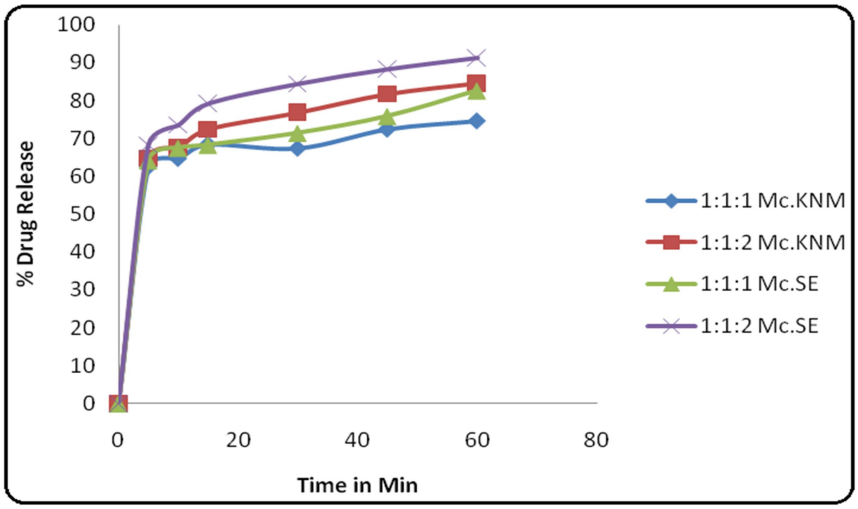

Figure 4: Dissolution studies of Etoricoxib-HP β cd- Citric acid complexes in phosphate buffer pH 6.8.

Table 11: Dissolution studies of Etoricoxib-Hp β- cd/citric acid/tartaric acid/pvpk30 by Freeze drying method

Time	Percentage drug release* ± SD		
	Citric acid 2M	Tartaric acid 2M	PVP k-30 2%
0	0	0	0
5	96.08±0.43	92.65±0.42	90.12±0.12
10	99.61±0.21	97.57±0.39	92.64±0.37
15	100.85±0.46	99.12±0.11	93.45±0.49
30	100.71±0.27	97.21±0.41	91.99±0.15
45	98.52±0.45	93.86±0.94	90.12±0.71
60	96.99±0.74	92.27±0.23	87.65±0.54

*= average cumulative drug release of triplicate samples (n=3)

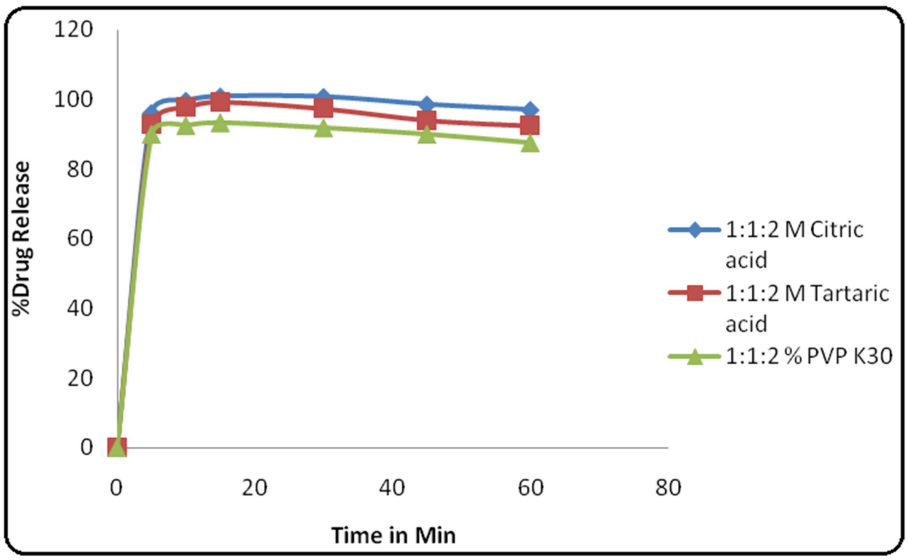

Figure 5: Dissolution studies of Etoricoxib-Hp β cd/citric acid/tartaric acid/pvpk30 by Freeze drying method

Table 12: Dissolution characteristics of optimized formulation and Innovator Tablet

Time in min	Percentage drug release* ± SD	
	F1	Innovator
5	90.57±0.54	89.35±0.38
10	98.32±0.52	93.72±0.44
15	100±0.57	96.67±0.41
30	100±0.98	98.52±1.09
45	100±0.37	100±0.74
60	100±0.57	100±0.68

*= average cumulative drug release of triplicate samples (n=3).

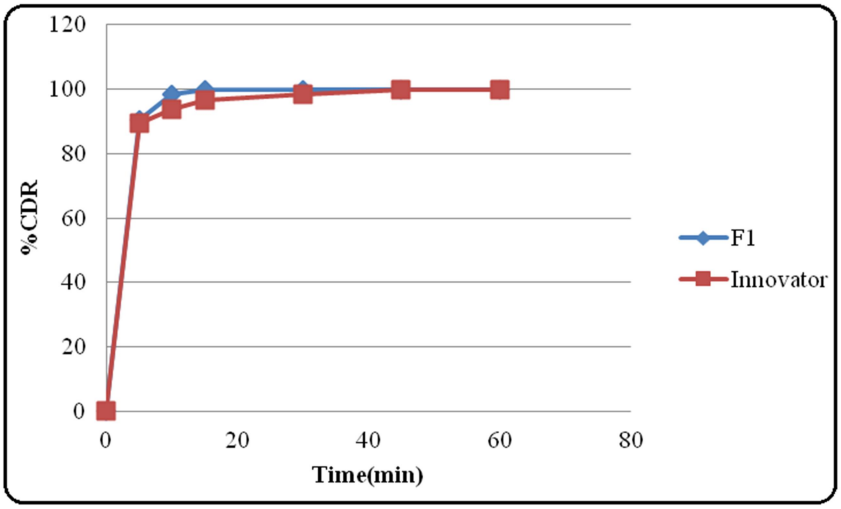

Figure 6: Comparison profiles of optimized formulation with innovator

Table 13: Comparison of release profile

Formulation trials	F2 value	F1 value	Influence of dissolution profile in comparison to innovators product as per FDA standards
F1	78.89	1.83	Similar

Stability studies

Table 14: Stability study for Optimized formulation (F1)

Evaluation Parameters	Initial	At 25°C ± 2°C / 60 % RH ± 5%	
		After 1 month	After 2 month
In vitro disintegration Time(s)	560	560.2	560.19
Hardness(kg/cm^2)	4.4±0.68	4.3	4.2
Drug content (%)	99.4±0.12	99.50±0.77	97.11±1.07

Table 15: Drug release profile of optimized formulation (F1)

Time (min)	Initial	% Drug release	
		At 25°C ± 2°C / 60 % RH ± 5%	
		After 1 month	After 2 month
0	0	0	0
5	90.57±0.54	90.1	89.71
10	98.32±0.52	98.01	97.53
15	100±0.57	100.25	99.89
30	100±0.98	100.45	100.01
45	100±0.37	100.33	99.98
60	100±0.57	100.26	100.11

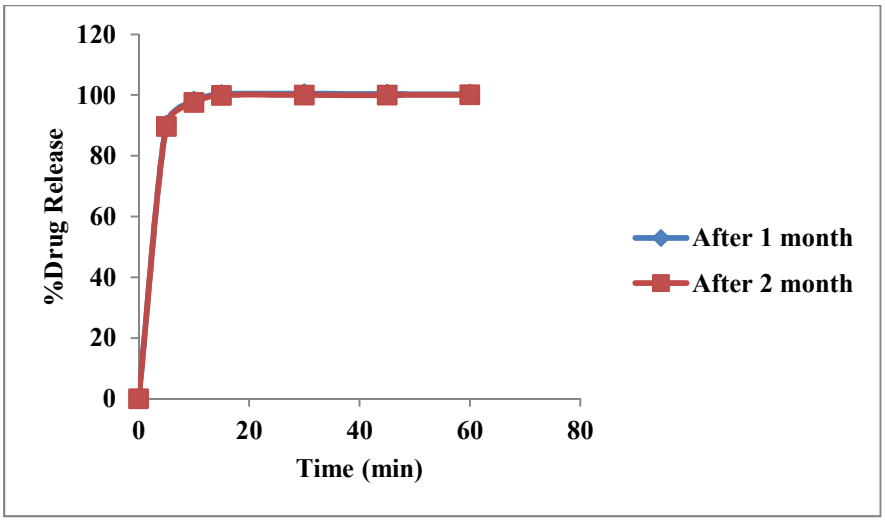

Fig 7: *In vitro* dissolution profile of stability batch after 1st and 2nd months at 25^0 C/ 60% RH

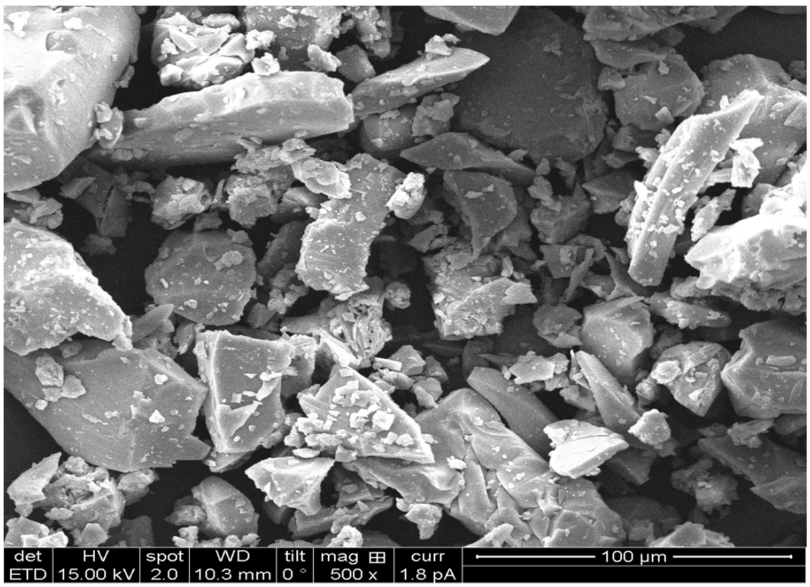

Figure 8: SEM of Etoricoxib- HP β cd- Citric acid (1:1:2M) freeze dried formulation

CONCLUSION

In the present investigation inclusion complexes of Etoricoxib were prepared with β cd, HP β cd. These systems are useful in enhancing aqueous solubility and hence oral bioavailability. Complexes prepared with HP β cd and Citric acid by freeze drying method has shown good release of drug, (100.85% at 15th min). Conventional tablets of etoricoxib was successfully prepared with etoricoxib-HP β cd- Citric acid (1:1:2M) complex using croscarmellose sodium. The formulation F1 was an optimized formulation which has similarity factor of 78.89 with that of marketed product.

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