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RESEARCH ARTICLE

An optimized commercially feasible milling technique for molecular encapsulation of meloxicam in β-cyclodextrin

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Abstract

Background: The practical applicability of solid dispersions (SD) for improvement of oral bioavailability of poorly water-soluble drugs has still remained limited because of lack of feasibility for scale-up of manufacturing processes. The present research work deals with the preparation of SDs of meloxicam (MLX) with β-cyclodextrin (β-CD) by the ball-milling technique to overcome the scale-up issues.

Methods: Phase-solubility studies were conducted to analyze the influence of β-CD on solubility of MLX. *In vitro* dissolution studies on various complexes as well as tablets prepared on pilot scale in an industrial set up were performed and compared with the marketed products. Physicochemical characterization of optimized complexes was done using various methods to study drug-β-CD interaction.

Results: Solubility of pure MLX in water at 25°C was found to be only 9.4 µg/mL. The AL type of phase-solubility profile of MLX with β-CD [stability constant ($K_{1:1}$) = 22.056 M^{-1} and Gibbs free energy (ΔF^o) = –7.665 KJ/mole] confirmed the solubility enhancement capability of β-CD. Milling time of 6 h was considered to be optimum and showed maximum enhancement of drug dissolution. The amorphous nature of the milled complex and mode of interaction of MLX with β-CD was confirmed by differential scanning calorimetry (DSC), x-ray diffractometry (XRD), Fourier transform infrared spectroscopy (FTIR) and proton nuclear magnetic resonance spectrophotometry (^{1}HNMR). Tablets containing MLX-β-CD (1:1.5 M) milled complexes showed the best release ($T_{90\%}$ = 10.94 min) compared to the marketed products ($T_{90\%}$ ≥ 450 min). Stability studies performed confirmed the integrity of the amorphous complex.

Conclusion: Stable inclusion complexes of MLX-β-CD with enhanced aqueous solubility and dissolution rate were prepared by a highly efficient and controlled large-scale milling technique.

Keywords: Ball-milling, tablets, industrial scale-up, solubility, dissolution

Introduction

Low dissolution rates related to poor water-solubility and fluctuations in plasma levels leading to erratic oral bioavailability are some of the well-known difficulties to be covered during the formulation development of existing and new drug entities[1–3]. According to the biopharmaceutics classification system, dissolution rate is the limiting factor for drug absorption of class II drugs[4]. An enhancement in dissolution rate is important to attain suitable and consistent blood-levels of these drugs[5,6]. Several practically viable methods like melting and solvent evaporation by co-precipitation, freeze-drying, spray-drying, etc. exist for the development of solid dispersions (SDs) of drugs in suitable carriers to enhance their *in vitro* and *in vivo* performances[7–11]. However, most of these technologies may be laborious, expensive and non-reproducible in terms of physicochemical characteristics. Those involving high temperatures and organic solvents may cause drug degradation, incomplete removal of the solvents and affect long-term stability of the drug in the vehicle. These drawbacks hinder the commercialization of most of the SDs designed today. In addition to the requirements of clinical efficacy and safety, the ability of the experimental formulation to be reproducibly manufactured on high-speed production equipment in a cost-effective manner is extremely important[12].

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The present research work explores the milling technique in order to overcome the problems mentioned above and at the same time enhance drug dissolution from SDs. A ball mill is a cylindrical device that rotates around a horizontal axis and is partially filled with the material to be ground plus the grinding medium. An internal cascading effect reduces the material to a fine powder, due to both impact and attrition effect. The grinding works on principle of critical speed, beyond which the balls start rotating along the direction of the cylindrical device; thus causing no further grinding. Industrial ball mills can operate continuously, fed at one end and discharged at the other end. Large to medium-sized ball mills are mechanically rotated on their axis, but small ones normally consist of a cylindrical capped container that sits on two drive shafts[13,14]. In SD prepared by milling technique, a fraction of the micronized drug might be molecularly dispersed in the matrix. When exposed to aqueous media, the carrier dissolves and the drug releases as fine colloidal particles. The resulting enhanced surface area produces higher dissolution rate and bioavailability of poorly water-soluble drugs[15,16].

Among the most popular carriers used in the formation of SDs are β-cyclodextrins (β-CDs). These are cyclic oligosaccharides with a remarkable ability to form inclusion complexes with various molecules that fit partially or entirely inside the β-CD cavity[17,18]. This phenomenon modifies the physicochemical properties such as solubility, dissolution rate and bioavailability of the guest molecules. From a pharmaceutical standpoint, β-CDs are economical, physiologically inert, bioadaptable and easily commercially available, and the drug-β-CD solid inclusion complex is convenient for oral administration[19–21]. Inclusion complexation of a number of drugs with β-CD using various techniques like kneading, co-precipitation, freeze-drying and spray-drying have been reported[22–25].

Drugs such as meloxicam (MLX), a non-steroidal anti-inflammatory oxicam derivative, having aqueous solubility in micromole/liter range are good candidates for solubility enhancement through β-CD complexation[26,27]. The solubility and dissolution rate of MLX in acid media are very poor which in turn not only affects its absorption and bioavailability but also leads to local high concentrations leading to gastrointestinal (GI) complications ranging from dyspepsia to fatal upper GI tract bleeding and perforation on chronic use[29]. The lipophilic inner cavity and the hydrophilic outer surface of β-CD is capable of interacting with MLX giving non-covalent inclusion complexes that improve aqueous solubility, dissolution rate and are capable of alleviating the undesirable properties of drug molecules through the formation of inclusion complexes[30,31]. Recently, many studies have investigated the effect of β-CD on improving MLX solubility but the available information on pilot scale-up of this subject is extremely limited, particularly by milling technique[32–34].

The aim of this study was thus to evaluate the enhancement effect of β-CD on MLX solubility and *in vitro* dissolution utilizing the milling technique. Possible physical interactions between the two components were investigated by performing Fourier transform infrared spectroscopy (FTIR), differential scanning calorimetry (DSC), x-ray powder diffraction (XRD) and proton nuclear magnetic resonance spectrophotometry (^{1}HNMR) studies[35–37]. The optimized MLX- β-CD SD was formulated into tablets, compared for *in vitro* dissolution with that of marketed conventional MLX tablet and subjected to accelerated stability studies.

Materials and methods

Materials

MLX was received as a gift sample by Unichem Lab. Ltd., Mumbai, Maharashtra, India. β-CD was generously donated by Cerestar (Indianapolis, IN). Conventional MLX tablets were purchased from a local Indian pharmacy (brand name undisclosed). Ultrapure water (Millipore; Agilent, Santa Clara, CA) and analytical grade reagents/chemicals were used throughout the study.

Phase-solubility studies

Phase-solubility studies were carried out according to the method reported in Higuchi and Connors[38]. Excess amount of drug (125 mg) was added in screw-capped conical flasks containing 50 mL of aqueous solution each of different concentrations (0, 0.125, 0.250, 0.375, 0.50, 0.625, 0.75 and 0.875 g) of β-CD in ultrapure water. The suspensions were continuously stirred on mechanical shaker (Siena equipments, Ahmedabad, Gujarat, India) at ambient temperature and 200 rpm for 48-h (this duration was previously tested to be sufficient to reach equilibrium). The suspensions were filtered through a 0.45 μ Millipore membrane filter. The filtrates were suitably diluted with water and analyzed, spectrophotometrically (Cecil 2000, UV/Vis spectrophotometer, England, Cambridge, England, UK), for the dissolved drug at 362 nm. Blank samples of β-CD at different concentrations used in the study were analyzed to rule out interference. All assays were performed in triplicate. The standard curve of MLX in water over a concentration range of 2–16 μg/mL at 362 nm was plotted. The mean calibration curve (regression equation: $y = 0.0531x + 0.003$) was found to be linear ($n = 6$) with a correlation coefficient of $r^2 = 0.9997$.

The data was treated statistically using linear least square regression and the apparent 1:1 ratio stability constant (K) and the Gibbs free energy (ΔF^o) were calculated from the phase-solubility diagram using Equations 1 and 2, respectively.

$$K = \frac{\text{Slope}}{y \text{ intercept } (1 - \text{slope})}, \quad (1)$$

where the y intercept corresponds to the intrinsic solubility of MLX in the absence of β-CD at 25 ± 1°C.

$$\triangle \mathrm{Fo} = -R\mathrm{T} \ln K \quad (2)$$

where, R is the ideal gas constant and K is the absolute temperature.

Preparation of SDs

The ball-milling technique was used to prepare inclusion complexes of MLX and β-CD in 1:1 w/w and 1:2 w/w ratio; as well as 1:1 mole (1:3.23 w/w) and 1:1.5 mole (1: 4.84 w/w) ratio. MLX and β-CD were separately sieved through 60#, weighed and mixed in geometric proportions in the abovementioned ratios. The porcelain ball mill (1.5 L capacity, 16 cm height, 11 cm internal diameter, Czecho origin, Ahmedabad, Gujarat, India) was charged first (up to 30% of its volume) with porcelain balls (2.2 cm average diameter, 16 g average weight) and then with the powder blend. Each mixture was ground at an optimum speed for different time intervals. The powder was scrapped sampled from the sides of the mill after every 2 h and analyzed for dissolution. The optimum milling time and speed for maximum dissolution was studied initially and was found to be 6 h and 45 rpm, respectively. The final milled product was stored in airtight containers till further analysis. Physical mixtures (PM) of MLX and β-CD were also prepared by mixing the powders in geometric proportions.

Drug content of SDs

Content analysis was performed using ultraviolet spectrophotometry. The mean calibration curve of MLX (regression equation: $y = 0.0536x - 0.0036$) in 0.1 N sodium hydroxide (NaOH) over a concentration range of 2–16 µg/mL at 365 nm was found to be linear ($n = 6$) with a correlation coefficient of $r^2 = 0.9999$ and hence could be employed for routine assay. Samples of PM and SDs prepared on lab scale and pilot scale, containing an equivalent of 15 mg of MLX were dispersed in a suitable quantity of 0.1 N NaOH and sonicated (Elma Transonic; 460/H, Kolpingstr, Singen, Germany) for 15 min. The filtered samples were suitably diluted with 0.1 N NaOH and measured for drug content.

In vitro dissolution studies

Pure MLX, PM and the milled SD's equivalent to 15 mg of MLX were subjected to dissolution studies in triplicate. Dissolution medium was 900 mL distilled water (containing 0.02% tween 80). The test was performed in a USP XXV Type II dissolution apparatus (Erweka DT 80; GmbH, Ottostraße, Heusenstamm, Germany). The stirring speed employed was 100 rpm and the temperature was maintained at 37°C ± 0.5°C. Aliquots of 5 mL were withdrawn at different time intervals, filtered and measured at 360 nm spectrophotometrically, after suitable dilution with the dissolution medium if needed, to determine the amount of drug released. The mean calibration curve of MLX (regression equation: $y = 0.0536x - 0.0036$) in the dissolution medium was found to be linear ($n = 6$) over a concentration range of 2–16 µg/mL with a correlation coefficient of $r^2 = 0.9995$.

DSC studies

The thermal behavior of MLX, β-CD, PM and optimized MLX-β-CD SD was studied using Perkin Elmer DSC 7 model (Perkin Elmer, Boston, MA) in aluminum pans under a nitrogen flow of 40 mL/min and heating rate of 10°C/min in a 20–250°C temperature range.

XRD studies

The XRD patterns were recorded using Philips X-ray generator (PW 1729) and automatic XRD model PW 1710 unit. The radiation used was Nickel filtered Cu $K_{\alpha 1}$ radiation having a wavelength of 1.542A°, operating at 35 KW and 20 mA in the range (2θ) of 5–60° at a scanning rate of 1.2°/min.

FTIR studies

The samples were subjected to FTIR spectroscopic studies by KBr disc method using JASCO FT/IR-5300 Spectrophotometer (CourtEaston, MD). The samples were scanned from 4000 to 400 cm^{-1} at room temperature.

^{1}HNMR studies

The ^{1}HNMR studies were performed with a Bruker FTNMR spectrophotometer, operating at 500 MHz using deuterated dimethyl sulfoxide (DMSO-d_6) as the solvent.

Tablet preparation

Tablets of MLX, MLX:β-CD (1:1.5 M) PM and MLX-β-CD (1:1.5 M) ball-milled SD were prepared by the wet granulation method using PVP K-30 in isopropyl alcohol as the binder. Starch 1500 and microcrystalline cellulose were added as internal disintegrant and diluent, respectively. The damp mass was sieved through 14# and the granules were tray-dried for 1 h at 60°C or till the moisture content reached 3.5%; which was determined on a Mettler Toledo moisture analyzer balance. The dried granules were then re-sieved through 20#. Sodium starch glycollate, talc and magnesium stearate were used as external disintegrant, antiadherent and glidant, respectively. The tablets were compressed using Cadmach single tablet press machine using 9-mm flat beveled edge punches to a final weight of 250 mg which was equivalent to 15 mg of MLX. The tablets were evaluated for various quality control parameters like hardness, weight variation, thickness, disintegration time and friability. The drug content and *in vitro* dissolution test in distilled water (containing 0.02% tween 80) as well as phosphate buffer pH 7.2 was compared with three different conventional marketed tablets.

Pilot scale batches

In a pilot plant, a formula is transformed into a viable, robust product by the development of a reliable and practical method of manufacture that effects the orderly

transition from laboratory to routine processing in a full-scale product facility. Scale-up batches of MLX, MLX:β-CD (1:1.5 M) PM and MLX-β-CD (1:1.5 M) milled complex were taken in Unichem Labs Ltd. Mumbai, Maharashtra, India. The batch size was 50,000 tablets for 15 mg dose and each batch was repeated in triplicate. The milled complexes were prepared by loading MLX and β-CD in a pilot scale ball mill (capacity = 8 L). The powder blend was milled for 6 h at a speed of 45 rpm, unloaded and sifted through 40# mechanical sieve. Granules were prepared using the technique and excipients mentioned above in a rapid mixer granulator (Mec-Well Pharma, Mumbai, Maharashtra, India). The tablets were compressed on a 16 station Cadmach rotary machine using 9-mm flat beveled edge punches to a final weight of 250 mg and evaluated for various quality control parameters.

Stability studies

The optimized scaled-up batches of MLX-β-CD milled complex (1:1.5 M) were blister packed and subjected to stability tests as per ICH guidelines at 30°C/60% relative humidity (RH) and 40°C/75% RH for 6 months and up to 12 months in ambient conditions.[39] Samples were withdrawn periodically and observed for any change in physicochemical parameters like visual appearance, mechanical properties, drug content and *in vitro* dissolution.

Statistical analysis

The dissolution data from different formulations was presented as mean ($n = 6$) and compared for statistical significance at $p<0.05$% level by one-way analysis of variance using SPSS 15.0 Software for Windows (SPSS Inc., Chicago, IL).

Results and discussion

Phase-solubility studies

The plot of drug solubility against increasing polychromatic x-ray microdiffraction concentrations investigated at 25 ± 1°C is represented in Figure 1. The solubility curve was classified as A_L type according to Higuchi and Connors[38]. Solubility of pure MLX in water at 25°C was found to be only 9.4 μg/mL. The extent of interaction between the drug and the carrier in aqueous media characterized by the apparent stability constant $K_{1:1}$, calculated according to the equation given by Higuchi and Connors, was found to be 22.056 M^{-1} whereas the Gibbs free energy (ΔF^o) was −7.665 KJ/mole[38]. The apparent stability constant and negative Gibbs energy confirmed the solubility enhancement of MLX by β-CD in aqueous state.

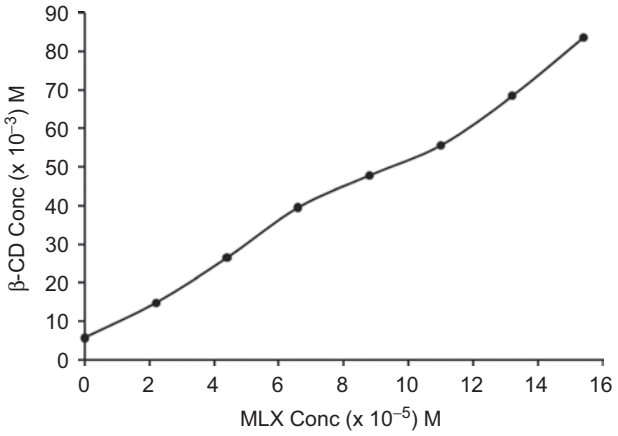

Figure 1. Phase-solubility plot of meloxicam (MLX) with β-cyclodextrin (β-CD).

Drug content of SDs

The drug content of the milled products manufactured at small scale ($n = 3$) and pilot scale ($n = 3$) and analyzed spectrophotometrically was found to be 99.113 (±0.552)% and 100.44 (±1.161)%, respectively of the theoretically added amount in the various dispersions.

Optimization of milling time

The influence of milling time on *in vitro* dissolution was studied on MLX and MLX-β-CD milled complexes at intervals of 2 h. Although it is believed that increasing the milling time should enhance the dissolution rate of drugs, it was observed that milling beyond 6 h had no significant effect on the dissolution of MLX ($p<0.05$). In fact, the $T_{25\%}$, $T_{50\%}$, $T_{75\%}$ and $T_{90\%}$ values of MLX-β-CD 1:1 w/w dispersions ball-milled beyond 6 h, were found to be higher than those milled for 6 h, however, the difference was not significant (Table 1). This could be attributed to the fact that increase in milling time leads to development of static charges, which may cause the particles to aggregate and hence retard the dissolution rate. The optimum milling time for all samples was therefore considered to be 6 h.

In vitro dissolution studies

The dissolution profiles of MLX, PM and various milled SDs are depicted in Figure 2. Tween 80 (0.02% w/v) was found to be optimum as a suspending agent to avoid powder floating on the surface of the dissolution medium. The initial dissolution rate of pure MLX was extremely slow and erratic with only 6.91(±1.17)% of the drug dissolved in 10 min and 18.02 (±1.39)% dissolution at the end of 1 h. This could be due to its highly hydrophobic nature and poor wettability. Rapid dissolution is a characteristic behavior observed for various SDs. It was noted that the ball-milled products of all MLX:β-CD ratios showed a faster release as compared to the pure drug and PM. The dissolution parameters of all the samples under study are listed in Table 1. The sequence of improved dissolution rate of the samples based on their $T_{90\%}$ values was found to be in the following order: MLX < 1:1 M PM < 1:1 w/w SD < 1:2 w/w SD < 1:1 M SD < 1:1.5 M SD. The improvement in the dissolution rate of the dispersed systems may be attributed to the decrease in degree of crystallinity of the active material due to milling and the surface acting property of the carrier which together attributed to the increase in both wettability and solubility of the drug.

Table 1. Dissolution parameters of various MLX-β-CD samples.

Sample	Time (min) $T_{25\%}$	$T_{50\%}$	$T_{75\%}$	$T_{90\%}$	First order rate constant	r
MLX	70.68	182.96	374.45	627.78	0.0016	0.9616
MLX (BM– 6 h*)	71.95	196.38	386.89	643.56	0.0019	0.9171
MLX:β-CD (1:1.5 M) PM	67.99	179.93	371.27	624.19	0.0015	0.9458
MLX:β-CD (1:1 w/w) BM– 6 h**	21.94	58.27	120.37	202.46	0.0049	0.9877
MLX:β-CD (1:1 w/w) BM– 12 h**	21.076	58.96	125.97	203.86	0.0051	0.9531
MLX:β-CD (1:1 w/w) BM– 18 h**	21.96	59.07	138.96	204.15	0.0199	0.9715
MLX:β-CD (1:1 w/w) BM– 24 h**	22.97	60.06	140.74	204.93	0.0074	0.9164
MLX:β-CD (1:2 w/w) BM– 6 h	11.75	38.71	64.78	145.71	0.0065	0.9825
MLX:β-CD (1:1 M) BM– 6 h	5.11	25.08	59.22	74.35	0.0086	0.9332
MLX:β-CD (1:1.5 M) BM– 6 h	2.56	7.59	16.19	27.53	0.0350	0.9842

*BM, ball-milled.
**Insignificant difference (p<0.05).
β-CD, β-cyclodextrin; MLX, meloxicam.

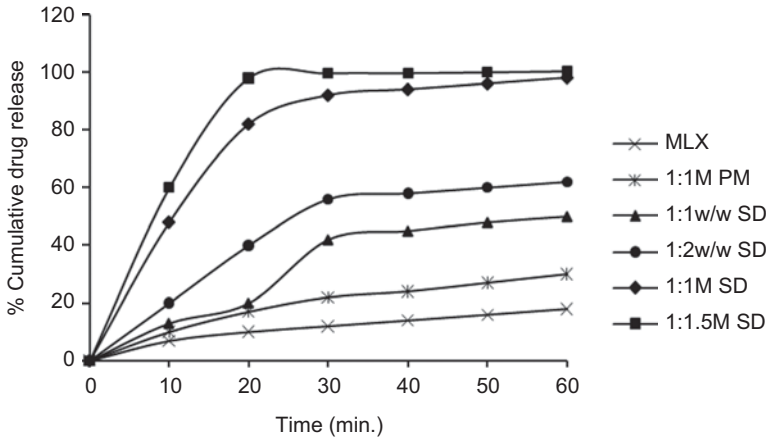

Figure 2. Dissolution profiles of various meloxicam (MLX) with β-cyclodextrin (β-CD) samples.

DSC studies

DSC enables quantitative detection of all processes in which energy is required or produced (i.e. endothermic and exothermic phase transformations). To characterize possible interactions between the drug and β-CD in the solid state, DSC thermograms of MLX, β-CD, MLX-β-CD PM and MLX-β-CD SD (1:1.5 M) were recorded (Figure 3). The DSC graph of pure MLX showed a sharp endothermic peak at 268.5°C, which is indicative of its melting temperature. The thermogram of β-CD depicts an endothermic peak at 67.9°C due to its dehydration process. The DSC pattern of MLX-β-CD PM, shows the presence of peaks of both the pure compounds, except with the difference that the drug-melting endotherm had slightly shifted from its original position of 268.5–262.5°C; and also the endotherm did not appear as a sharp peak. This slight shift could be due to a weak interaction between the drug and β-CD. The thermogram of MLX-β-CD SD (1:1.5 M) prepared by milling method exhibited almost complete disappearance of the endothermic peak characteristic of MLX; which can be attributed to its amorphous character in the fused state; strongly indicating that the drug is well dispersed in β-CD matrix and its re-crystallization is restrained. Based on the percentage peak areas calculated from the endothermic peak of MLX, from the thermograms of pure drug and drug-β-CD SD, the complexation was found to be 93.94 (±1.27)%. The results of thermal analysis are thus suggestive of maximal complex formation and dissolution of the drug in the molten carrier.

XRD studies

The XRD pattern of MLX and β-CD (Figure 4) showed peaks that were intense and sharp; indicating their crystalline nature while that of the PM was found to be a combination of the drug and β-CD, with little decrease in the peak intensity. However, the XRD pattern of the MLX-β-CD complex was found to be diffused and different. Crystallinity was determined by comparing some representative peak heights in the diffraction patterns of the binary systems with those of MLX. The characteristic

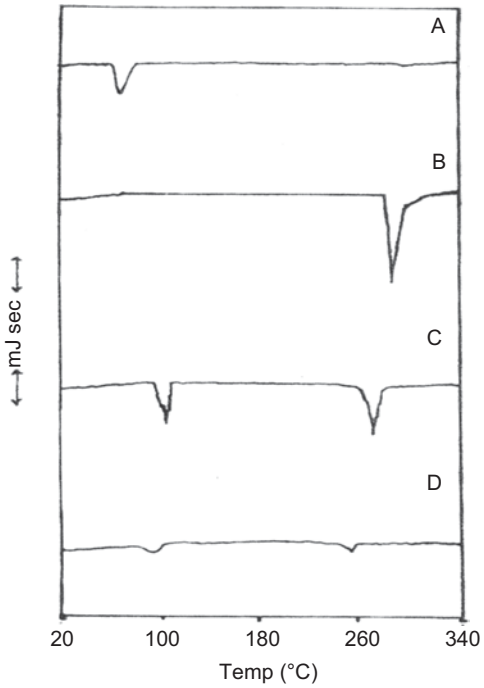

Figure 3. Differential scanning calorimetry (DSC) thermograms of various meloxicam (MLX) with β-cyclodextrin (β-CD) samples.

peaks at 15.5°, 17.5°, 22°, 23°, 24.5° and 28° (2θ) observed in Figure 4A for MLX are significantly diminished in the complex (Figure 4B). The relative degree of crystallinity (RDC) was calculated using the equation $RDC = H_{sample}/H_{ref}$; where, H_{sample} is the peak height of the MLX-β-CD complex and H_{ref} is the peak height at the same angle for MLX. The characteristic peak intensity ratios are listed in Table 2. The RDC values of the complex at 2.5° and 15° (2θ) were 0.1026 and 0.1842, respectively thus confirming the formation of a new solid phase for the milled complex.

FTIR studies

The interaction between the drug and its carrier often leads to identifiable changes in the IR profile of the SD. FTIR spectral studies were employed to confirm complex formation of MLX with β-CD. The spectra of the SD was compared with that of MLX, β-CD and the corresponding PM (Figure 5). The intense peaks appearing in the spectra of MLX and β-CD are due to the asymmetric stretching vibrations of their functional groups. The IR spectrum of β-CD (Figure 5A) shows prominent peaks at 3389.64 cm^{-1}(O–H), 2924.86 cm^{-1} (C–H), 1649.90 cm^{-1} (H–O–H bending), 1157.71 cm^{-1} (C–O) and 1028.51 cm^{-1} (C–O–C). The IR spectrum of pure MLX (Figure 5B) showed characteristic principle peaks at 1529.10 cm^{-1} (aromatic–C-C-), 3085.49 cm^{-1} (aromatic–C–H–), 2930.11 cm^{-1} (C–H aliphatic), as well as at 1346.43 cm^{-1} (–S=O), 3292.79 cm^{-1} (–S–N–) and 1620.35 cm^{-1} (–N–H–). The spectrum of the PM (Figure 5C) showed a summation

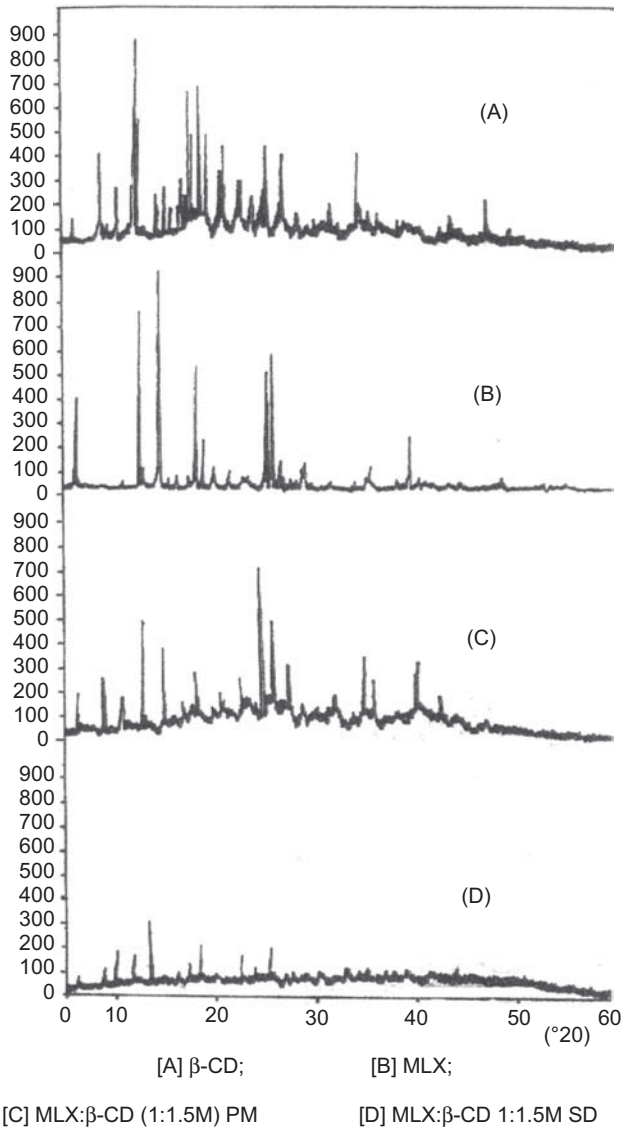

Figure 4. X-ray diffractograms of meloxicam (MLX) with β-cyclodextrin (β-CD) samples.

Table 2. Shifts in XRD peaks of MLX after complexation.

	Peak intensity		
2θ	MLX	MLX:β-CD (1:1.5 M)	RDC*
2.5°	390	40	0.1026
13°	760	340	0.4474
15°	950	175	0.1842
26°	575	230	0.4000

*RDC, relative degree of crystallinity.
β-CD, β-cyclodextrin; MLX, meloxicam; XRD, x-ray diffractometry.

effect i.e. simple superposition of the peaks due to the functional groups of the two compounds, indicating the presence of MLX in crystalline state. In the spectrum of MLX-β-CD complex (Figure 5D), the presence and absence of characteristic peaks associated with specific structural characteristics of the drug molecule were noted, however; there were no new peaks, indicating any new chemical bond formation between the two in solid state.

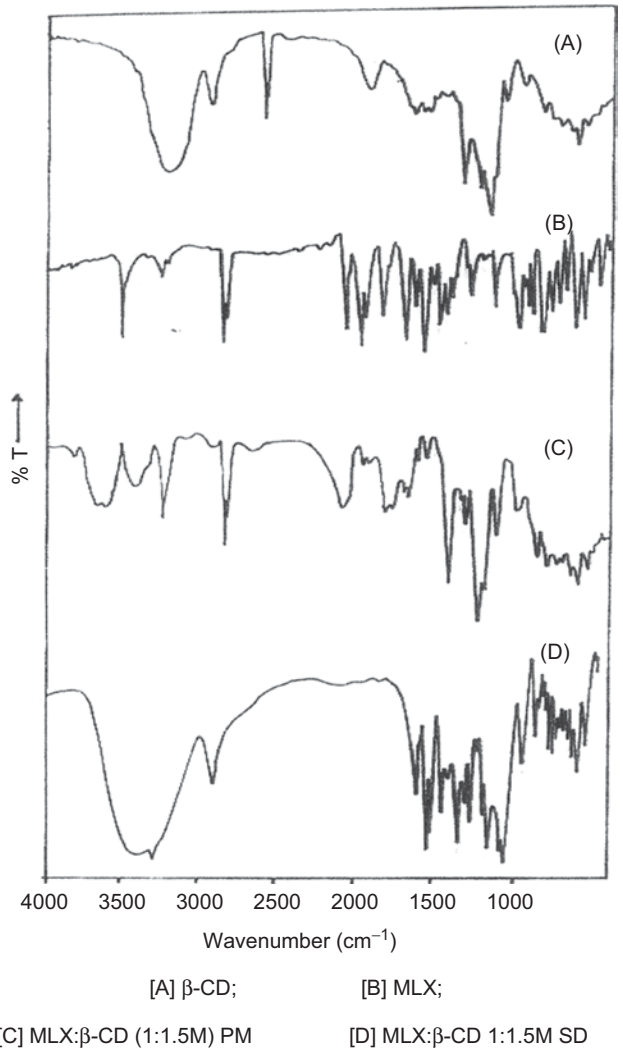

Figure 5. Fourier transform infrared spectroscopy (FTIR) spectra of meloxicam (MLX) with β-cyclodextrin (β-CD) samples.

The spectrum of the complex showed appearance of an intense broad peak at 3429.7 cm^{-1}. This peak broadening indicates possible hydrogen bonding between MLX and β-CD. Shifts are seen in the peak of the aromatic–C–H– (2925.6 cm^{-1}) and aromatic–C-C- (1569.0 cm^{-1}) stretching of the benzene ring, suggesting that these groups are taking part in hydrogen bonding leading to entrapment of the aromatic ring of the guest molecule in the hydrophobic cavity of the host (as represented in Figure 6), whereas the sulfide (1352.3 cm^{-1}) and amide group (1627.2 cm^{-1}) peaks do not exhibit any significant contribution in the hydrogen bonding process. These chemical shifts could be attributed to the physical interaction of the drug with β-CD which in turn enhance wettability, aqueous solubility and dissolution of the drug.

^{1}HNMR studies

The inclusion of MLX in β-CD cavity is confirmed from the change in chemical shift of some of the guest and host protons in comparison with the chemical shifts of the same protons in the pure form (Figure 7). The phenomenon results in alteration of chemical shift values

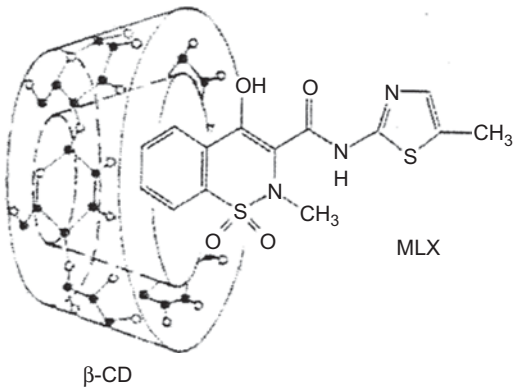

Figure 6. Proposed schematic representation of inclusion phenomenon of meloxicam (MLX) in β-cyclodextrin (β-CD).

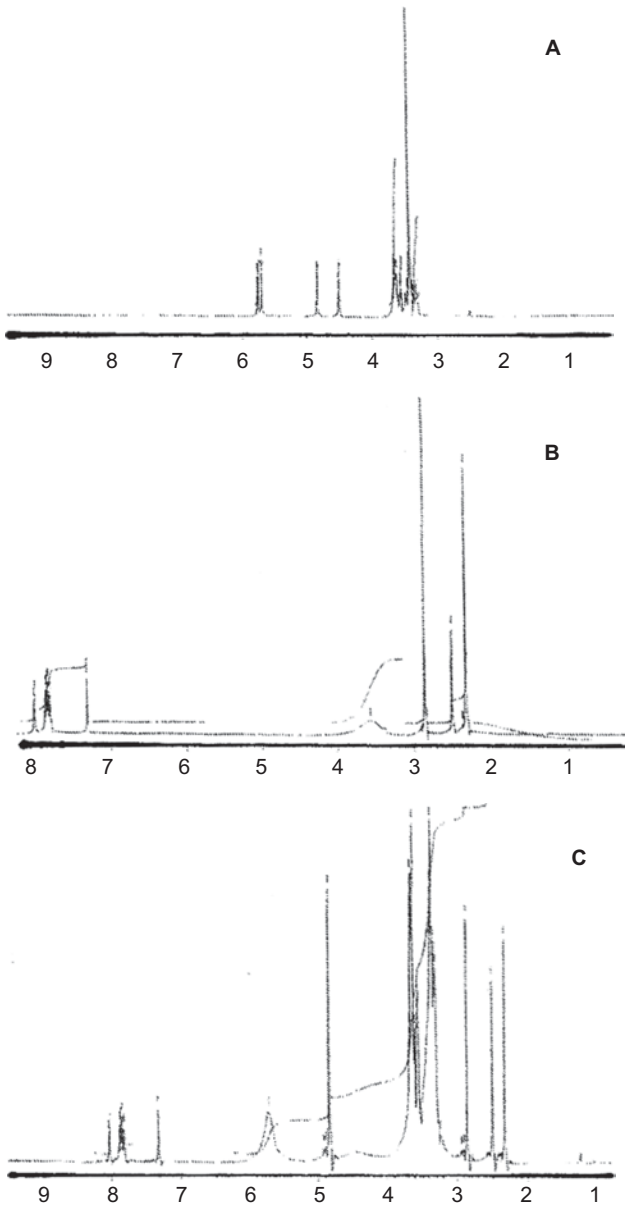

Figure 7. Proton nuclear magnetic resonance spectrophotometry (^{1}HNMR) spectra of (A) β-cyclodextrin (β-CD), (B) meloxicam (MLX) and (C) MLX-β-CD solid dispersions (SD).

for most aromatic protons of the drug supporting the view that at least one of the aromatic ring interacts with the inner protons of β-CD by forming hydrogen bonds and through Vander Waals forces of attraction. It is evident from the graphical representation in Figure 8 that the hydrogen atoms of the MLX benzene ring (C^1_2– C^1_6) show a significant down field shift on complexation with β-CD indicating its entrapment inside the β-CD cavity resulting in complex formation. In addition, the inclusion phenomenon resulted in high up-field shift changes in the β-CD protons situated inside the cavity, (namely H-3′and H-5′) while insignificant shift changes were observed for other β-CD protons. The entry of the aromatic ring should be more favorable through the wider rim (lined with primary hydroxyl groups) of the doughnut-shaped molecule as compared to the narrower rim (lined with the secondary hydroxyl groups) for stearic reasons. This assumption can be confirmed by performing more sophisticated NMR studies like rotating frame overhause effect spectroscopy under spin-lock conditions. The presented NMR data for the samples are in complete accordance with the FTIR results and confirm inclusion of the aromatic ring of MLX within the non-polar β-CD cavity.

Tablet formulation in lab and pilot scale

A series of tablet batches of MLX, MLX:β-CD (1:1.5 M) PM and MLX-β-CD (1:1.5 M), each containing 15 mg of the drug were prepared in triplicate on a lab scale followed by pilot scale batches. The technological properties and drug content for each batch composition of the examined tablet formulations at pilot scale are reported in Table 3. The pilot scale study was planned and conducted only after achieving satisfactory results at the lab scale level; hence, it was considered unnecessary to tabulate the quality control results of the lab batches also. All the batches showed homogeneous, consistent and satisfying technological properties, and no considerable differences were noticed when the batches were repeated. Incorporation of super disintegrants like starch 1500 and sodium starch glycollate during granulation as well as post-granulation, respectively lead to rapid disintegration of tablets in less than 2 min for all the batches making the dispersed granules available for faster dissolution. The use of PVP K-30 as a binder in the wet granulation process further enhanced the dissolution of the drug in tablet form compared to its intrinsic solubility due to its wetting properties which is very well reported by many authors. The dissolution profiles of the formulated tablet batches were compared to that of three different marketed products and are mentioned in Table 4. It is evident, that a significant improvement in drug dissolution profile was achieved from the ball-milled MLX-β-CD (1:1.5 M) complex tablets with respect to the plain drug and the corresponding PM, because of the wetting, solubilizing and amorphizing properties of the carrier towards the drug. The time taken for 90% of the drug to dissolve ($T_{90\%}$) from the milled complex tablets was only 10.94 min, whereas for the marketed tablets it ranged from 7 to 8 h. As the drug shows pH-dependent solubility it was considered of interest to compare the dissolution profile of the pilot scale complex with that of the marketed products. It was observed that none of the marketed products showed a release of more than 50% whereas the milled tablets showed 100% drug release at the end of 10 min. The milling technique can therefore be considered as a promising technique for effective interaction between drug and CD and can be used extensively for preparation of inclusion complexes to overcome solubility problems, improve drug dissolution

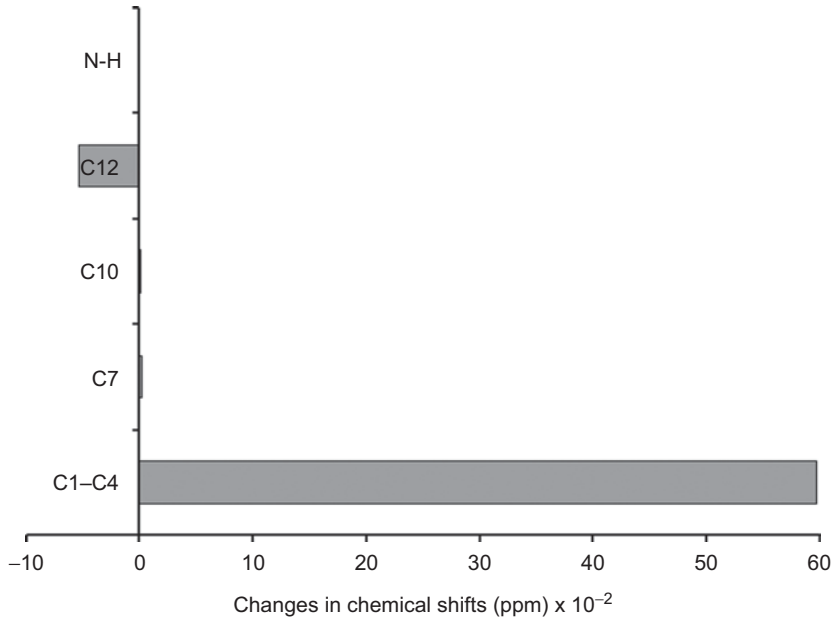

Figure 8. Chemical shift changes in proton nuclear magnetic resonance spectrophotometry (^{1}HNMR) signals induced by complexation of meloxicam (MLX) in β-cyclodextrin (β-CD).

Table 3. Technological properties and drug content of different pilot scale batches ($n=3$) of MLX.

	Formulation		
Parameter	MLX tablets	MLX:β-CD (1:1.5 M) PM tablets	MLX:β-CD (1:1.5 M) milled complex tablets
Mean weight (mg)††	251.31 ± 2.68	249.88 ± 1.53	250.64 ± 2.07
Hardness (kg/cc)†	4.0 ± 0.5	3.5 ± 0.25	4.0 ± 0.5
Thickness (mm)†	3.11 ± 0.065	3.07 ± 0.091	3.1 ± 0.044
Disintegration time (min)†	2.58 ± 0.84	1.54 ± 0.47	1.13 ± 0.25
Friability (% w/w)††	0.313	0.129	0.194
% Drug content††	99.87 ± 2.17	101.65 ± 1.93	100.29 ± 1.42

†$n=6$ tablets.
††$n=20$.
β-CD, β-cyclodextrin; MLX, meloxicam; PM, physical mixtures.

Table 4. Dissolution parameters of pilot scale MLX-β-CD and marketed tablets.

	Time (min)*				First order rate	
Tablet batch	$T_{25\%}$	$T_{50\%}$	$T_{75\%}$	$T_{90\%}$	constant	r
MLX*	81.95	165.93	297.25	538.71	0.0062	0.9972
PM (1:1.5 M)*	61.56	143.83	266.95	521.82	0.0025	0.9926
BM (1:1.5 M)	1.74	4.84	8.03	10.94	0.0095	0.9993
MKTD– 1*	75.94	163.48	329.34	506.35	0.00854	0.9935
MKTD– 2*	62.57	158.94	327.94	481.97	0.00467	0.9479
MKTD– 3*	50.95	139.95	302.35	445.54	0.00677	0.9834

*Significant difference ($p<0.05$) compared to BM (1:1.5 M).
β-CD, β-cyclodextrin; BM, ball-milled for 6 h; MKTD, marketed tablets; MLX, meloxicam; PM, physical mixtures

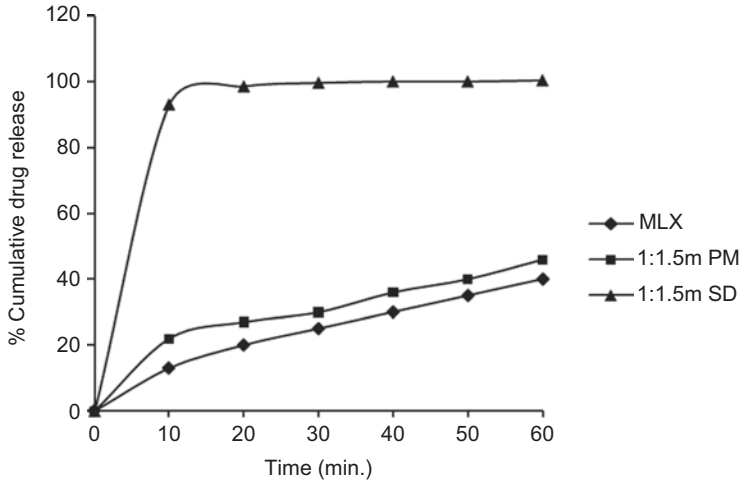

Figure 9. Dissolution profile of various pilot scale meloxicam (MLX) tablets.

and subsequently the absorption performance after oral administration.

Stability studies

The milled MLX-β-CD (1:1.5 M) complex tablets subjected to accelerated stability conditions exhibited good stability with respect to drug concentration, *in vitro* dissolution, mechanical properties and other physical attributes like size, shape, color, surface texture or any other visible flaws. The assay values for the samples stored under ambient conditions, 30°C/60% RH as well as 40°C/75% RH were found to range between 99.47% and 98.63% after 1, 2, 3 and 6 months of storage and differences were not statistically significant compared to the zero time assay results. In addition, there was no significant difference observed in the release profiles, conforming the stability and integrity of the amorphous complex even under accelerated conditions. The unaltered content analysis and dissolution data confirmed the stability of the amorphous nature of the milled complexes of MLX and β-CD and hence it was not considered essential to repeat the molecular characterization studies at the end of the stability test period.

Conclusions

SD of MLX with β-CD by an industrially feasible milling technique was achieved successfully and the amorphous

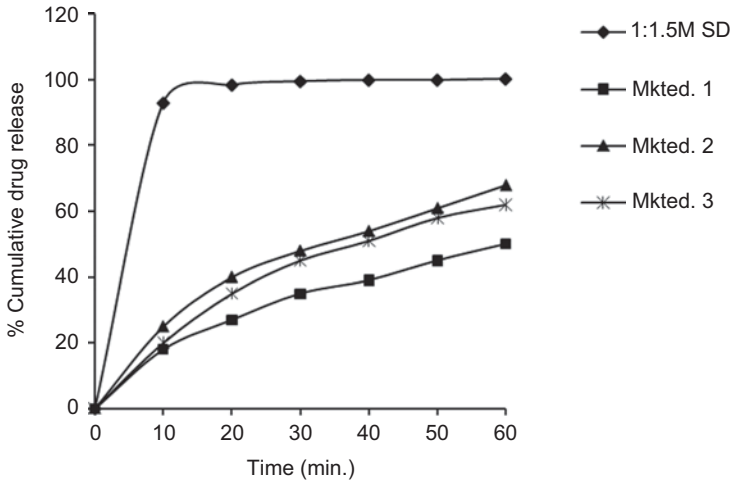

Figure 10. Comparison of dissolution profile of pilot scale meloxicam (MLX) with β-cyclodextrin (β-CD) milled complex (1:1.5 M) tablets and three different marketed tablets in distilled water (0.02% tween 80).

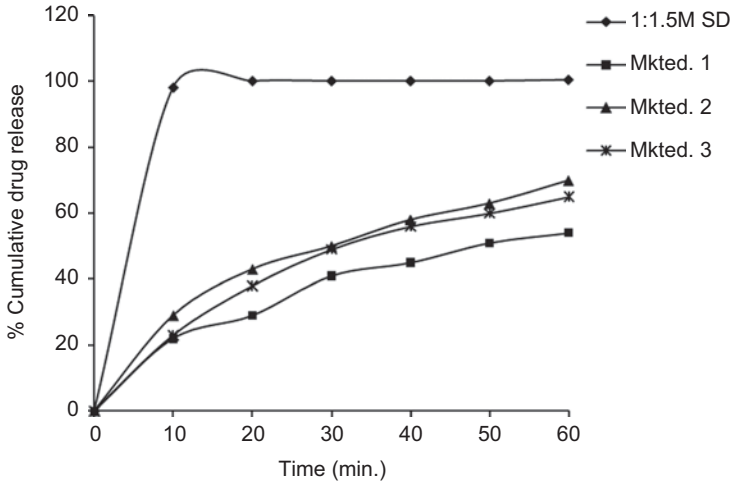

Figure 11. Comparison of dissolution profile of pilot scale meloxicam (MLX) with β-cyclodextrin (β-CD) milled complex (1:1.5 M) tablets and three different marketed tablets in phosphate buffer (pH 7.4).

nature of the complex was confirmed by molecular characterization. Tablets prepared by the wet granulation method exhibited good technological properties and improved dissolution profile allowing almost 100% *in vitro* release in first 10 min, with respect to the plain drug and marketed tablet formulations. The absorption and bioavailability of most of the hydrophobic drugs administered orally is directly related to its aqueous solubility and dissolution behavior. The satisfactory performance of this commercially scaled-up complexation method offers the triple advantage of improved gastrointestinal tolerability, better efficacy profile and faster onset of action.

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Declaration of interest

The authors report no conflicts of interest.

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