Formation of fine and encapsulated mefenamic acid form I particles for dissolution improvement via electrospray method


To cite this article: Nurul Karimah Zolkepali, Noor Fitrah Abu Bakar, M. Nazli Naim, Nornizar Anuar, Nurul Fadhilah Kamalul Aripin, Mohd Rushdi Abu Bakar, I. Wuled Lenggoro & Hidehiro Kamiya (2016): Formation of fine and encapsulated mefenamic acid form I particles for dissolution improvement via electrospray method, Particulate Science and Technology, DOI: 10.1080/02726351.2016.1246496

To link to this article: http://dx.doi.org/10.1080/02726351.2016.1246496
Formation of fine and encapsulated mefenamic acid form I particles for dissolution improvement via electrospray method


© 2016 Taylor & Francis

ABSTRACT
The potential of using electrostatic atomizer or electrospray in producing fine and encapsulated particle of mefenamic acid (MA) form I with β-cyclodextrin (βCD) was demonstrated in this study. Encapsulated MA-βCD with a molar ratio of 1:2 was prepared in water-ethanol suspension, followed by the electrospray process to atomize the droplet into fine dried particles. The working distance (WD) between the electrospray needle tip and the substrate were varied from 15 to 25 cm. The sizes of encapsulated MA-βCD particles were found to decrease from 91 ± 26 to 42 ± 35 nm as the WD increased. The dissolution rate of encapsulated particles of MA-βCD was found to be higher compared to the particles of as-received MA and the unencapsulated MA. The presence of the encapsulated MA-βCD was proven by a thermal analysis with the disappearance of MA peak after the atomization process. The x-ray diffraction analysis showed that the encapsulation occurred with the existence of new solid phase that was expected from interaction between MA and βCD and the appearance of C=C. Further analysis by transmission electron microscopy showed the size and morphology of MA-βCD particles when immersed in water and acetone. Encapsulated MA-βCD particles were solubilized in water but suspended as spherical shape in acetone.

1. Introduction
The dissolution rate of pharmaceuticals is an important aspect in drug development. In the market, approximately 40% of oral drugs are categorized as practically insoluble or poorly soluble in water. The limited dissolution rate arising from low solubility frequently results in low bioavailability of drugs with aqueous solubility lower than 100 µg/mL and generally presents dissolution-limited absorption (Takagi et al. 2006). Particularly for mefenamic acid (MA), its oral bioavailability is very low, probably due to poor solubility in water and insufficient dissolution rate (Shinkuma et al. 1984). Dissolution improvement of drugs remains a major challenge in the development of new drug treatments. Various approaches have been developed focusing on the improvement of the dissolution rate of poorly water-soluble drugs such as surface area enhancement through particle size reduction of solid compound (Tong et al. 2001; Enayati et al. 2011) and/or by drugs’ physicochemical properties manipulation through complex formation of the drug and polymer for encapsulation process (Klein, Polheim, and Viernstein 2000) by using β-cyclodextrin (βCD) (Corrigan and Stanley 1982). Srijamornskak et al. (2015) investigated the self-emulsifying formulation of MA composing of oil, surfactant, and co-surfactant for dissolution improvement. However, to the best of the authors’ knowledge, none of the works reported on the encapsulation of MA drug.

Encapsulation is a process of capturing solid particles, liquid droplets, or gaseous bubbles as a central material in a solid or liquid envelope (shell) made of another material. Encapsulation serves as an advantage for drug availability in which it controls the release of drug. The hydrophilic outer surface material of the encapsulated drugs forms hydrophilic chains with the aqueous media and hence aids the drug dissolution process (Yeo, Gagnon, and Chang 2005; El, Pierre, and Thorstein 2013; Sheng et al. 2014). For the encapsulation of drug, complex formation of cyclodextrin has been studied and has been used in various pharmaceuticals for drug dissolution improvements such as encapsulation formation between etodolac, which is practically an insoluble drug with βCD (Ammar et al. 2013), and encapsulation between isatoic anhydride and βCD (Patil et al. 2013).

There are several common methods available for particle size reduction or encapsulation process such as spray and freeze dry methods. However, these methods have their own limitations. Spray drying (Zheng and Chow 2009) involves elevated temperature during the process, whilst freeze drying (Zhang et al. 2011) requires low temperature during the solidification process. Excessive heat used during spray drying...
may degrade the compounds and slow freezing might cause irreversible crystal formation. As an alternative, electrospray process can be used to produce encapsulation of drugs at room temperature and has been successfully carried out by previous researchers like Bohr et al. (2011), who made an attempt to produce PLGA microparticles loaded with celecoxib by electrospray and fabricated particles by electrospray process for bioproduction between alginate and chitosan microcapsules (Fukui et al. 2010).

The effectiveness of encapsulation process can be increased by inducing some electrical forces during the process, as used in electrospray or also known as electrohydrodynamic atomization process. The electrospray mechanism for the formation of fine particles involves five major processes: (1) generation of charged droplet, (2) shrinkage of the droplets due to the solvent evaporation, (3) repeated disintegration of the droplets due to Coulomb fission, (4) formation of dry particles, and (5) deposition or collection of particles (Naim et al. 2010). Droplet fission and solvent evaporation used during the process encapsulate the drug and solidify the shell material, (Jaworek and Sobczyk 2008) whilst aiding the particle size reduction (Nemes, Marginean, and Vertes 2007).

One of the advantages of electrospray is its capability to control parameters such as deposited distance (working distance (WD)), properties of solution, nozzle diameter, and electric field (Ganan-Calvo 1994). Emission of liquid suspension from the jet in the electrospray disintegrates the droplets and results in the droplet’s shrinkage associated with the encapsulation of the drugs with hydrophilic matrix during solvent evaporation process. During this process, the disintegrated droplet experiences interfacial tension stress and the electrostatic forces will overcome the surface tension that leads to liquid breakup under evaporation (Ambrus et al. 2013). As the mass of droplets reduces continuously, the repulsion of charge on the surface increases until it approaches the Rayleigh limit. At this stage, the droplets become unstable and cause the droplets to disrupt into smaller droplets than the primary droplets. During the outflow of droplets to the substrates, the charge on the droplets prevents aggregation, which results in the formation of monodisperse particles and deposit on the substrates (Wang et al. 2012).

In this work, we propose the use of electrospray process to encapsulate MA using βCD, with the intention to improve the dissolution of form I polymorphic form of MA, which is known to have a low solubility in aqueous solution.

### 2. Experimental procedure

#### 2.1 Materials and preparation of feed suspension and solution

Mefenamic acid, 2-(2,3-(dimethylphenyl) amino) benzoic acid form I (C₁₃H₁₅NO₂, MW = 241.29 g mol⁻¹, purity = 99%), was used without further purification. β-Cyclodextrin ((α-1,4)-linked α-d-glucopyranose) (C₆₆H₁₀₁O₃₀, MW = 1134.98 g mol⁻¹, purity = 99%) was selected as the encapsulation material and purchased from Sigma-Aldrich.

In this work, three types of particles were produced in the electrospray experiment, i.e., unencapsulated MA particles at two concentrations and encapsulated MA-βCD particles. Both the unencapsulated MA produced served as comparisons to the encapsulated MA-βCD. Prior to the electrospray process, a suspension of MA-βCD from two separate solutions was prepared. First, the MA solution was prepared by dissolving 24.2 mg of MA in 5 mL of ethanol. Then, the βCD solution was prepared by dissolving 227 mg of βCD in 20 mL of distilled water. Both solutions were stirred to ensure all particles were completely dissolved in their respective solvents. Then, the MA solution was added to the βCD solution drop by drop to form a suspension (Figure 1), while stirred for 30 min, with a molar ratio of 1:2 (MA solution to βCD solution). This molar ratio was used to ensure the βCD molecules encapsulated all the MA molecules, during the electrospray process. To determine the most stable suspension, the mixed solution was titrated with 0.1 M NaOH and 0.1 M HCl to obtain the suspension with the range of pH between 2 and 11. The suspensions were then measured using Zetasizer Nano ZS instrument (Malvern Instrument Ltd., Worcestershire) for the zeta potential values.

For the solution of unencapsulated MA, the MA solutions in acetone were prepared with different concentrations. The MA (as-received) was dissolved in acetone with the percentage of MA mass to acetone volume of 0.33 wt./vol% (3.33 mg/mL) and 0.2 wt./vol% (2 mg/mL), respectively. Then, the solutions were stirred for 30 min at room temperature to ensure all particles were completely dissolved.

The electrical conductivity, which is to be used in Equations (1)–(3) for primary droplet estimation, was measured using a conductivity meter (Mettler Toledo, Tokyo).
2.2 Electrospraying of MA-βCD suspension and MA solutions

Atomization of MA-βCD suspension and MA solutions was performed using an electrospray system as shown in Figure 2. The prepared MA-βCD suspension was filled in a 20 mL plastic syringe. The syringe was attached to a stainless steel needle, which acted as a nozzle. The length of the nozzle was 38.1 mm, with inner and outer diameters of 0.15 and 0.30 mm, respectively. The prepared MA-βCD suspension was pumped through a 0.15 mm inner diameter of needle, and the flow was controlled by the infusion of syringe pump (NE-1000, New Era Pump Systems Inc., New York). Electric field was applied to both the needle tip and counter electrode with positive and negative polarity, respectively. A counter electrode of 7 mm diameter with a circular opening in the center was positioned perpendicularly to the needle. The electrospray was performed under cone-jet mode in a closed chamber. A digital camera equipped with a macro lens was used to observe the electrospray modes. The same procedures were repeated for MA solution to produce unencapsulated MA particles. The grounded aluminum foil with the size of 10 × 10 cm was used as a substrate to collect the deposited particles sprayed from the electrospray. For the MA-βCD suspension, the distance from the needle tip to the substrate (known as WD) was varied in the range of 15–25 cm, whilst for the MA solutions, the WD used was varied in the range of 5–15 cm. The WD for deposition of encapsulated MA-βCD varied from 15 to 25 cm while for production of unencapsulated MA varied from 5 to 15 cm to ensure droplet fission occurred before deposition of particles to the substrate due to different solvents used. Then, the deposited particles were collected from the substrate and dried at 50°C for 30 min and kept in a desiccator for further analysis. The electrospray parameters used in this study are summarized in Table 1.

2.3 Prediction of primary droplet size of MA-βCD suspension

Scaling laws were used to estimate the size of primary droplet produced by the cone-jet mode of the electrospray, and in this

![Figure 2. Electrospray experiment set-up and the mechanism of the formation of solid encapsulated MA-βCD from MA-βCD suspension.](image-url)
work three models were used and compared for the sizes of the droplet, as follows:

Hartman et al. (2000):

$$d_d = \left( \frac{\rho \varepsilon_0 Q^3}{\gamma K} \right)^{1/3}$$  \hspace{1cm} (1)$$

Fernandez de La Mora and Loscertales (1994):

$$d_d = 1.66 \varepsilon_t^{-1/6} \left( \frac{Q \varepsilon_0 \varepsilon_\infty}{K} \right)^{1/3}$$  \hspace{1cm} (2)$$

Ganan-Calvo (1999):

$$d_d = 1.2164 \left( \frac{Q \varepsilon_0 \varepsilon_\infty}{K} \right)^{1/3}$$  \hspace{1cm} (3)$$

where \(d_d\), \(\rho\), \(\varepsilon_0\), \(Q\), \(\gamma\), \(K\), and \(\varepsilon_t\) represent the diameter of droplet, density of liquid (\(\rho = 1000 \text{ kg/m}^3\)), vacuum of permittivity \((8.8 \times 10^{-12} \text{ C}^2/\text{N/m}^2)\), flow rate of liquid (\(m^3/s\)), surface tension of liquid (\(\gamma = 0.04 \text{ N/m}\)), conductivity of the solution (S/m), and relative permittivity (dimensionless number with magnitude of 78.36), respectively.

Two main parameters that control the droplet size are the flow rate and the electrical conductivity of the solution. Therefore, to estimate the droplet size, the flow rate of the liquid was determined experimentally based on the stable cone-jet formation and the solution conductivity was measured using a conductivity meter.

Both theoretical and experimental numbers of droplets fission can be determined from the ratio of the volume of the primary droplets to the volume of the particles, which is assumed spherical. The volume of the primary droplets was calculated using the scaling law [Equations (1)–(3)]. Then, the theoretical values of the volume of the spherical particles were calculated from Hogan et al. (2007) using Equation (4), i.e., based on the diameter of the particles with the assumption that the droplet evaporation occurring during electrospray is without mass loss:

$$d_p = d_d \phi^{1/3}$$  \hspace{1cm} (4)$$

where \(d_p\) is the diameter of the particle produced by electrospray, and \(\phi\) is the volume fraction of the material in the solution.

Assume the shape of the particles on the substrate is a sphere. Therefore, the number of fission was calculated by dividing Equations (5) and (6), which is simplified into Equation (7):

$$\text{Volume of primary droplet} = \frac{4}{3} \pi \left( \frac{d_d}{2} \right)^3$$  \hspace{1cm} (5)$$

$$\text{The number of fission} = \left( \frac{d_d}{d_p} \right)^3$$  \hspace{1cm} (7)$$

2.4 Evaluation of as-received materials and electrosprayed products

The materials and products from the experiment were characterized using field emission scanning electron microscopy (FESEM) using an accelerating voltage of 10 kV. Prior to the analysis, all samples were sputter-coated with platinum under vacuum to avoid the charging effect during FESEM observation. The total numbers of 250 MA-βCD particles were selected to determine the average size that was observed using ImageJ software. On the other hand, for the experimental values, the volume of the spherical particles was calculated from FESEM (JEOL JSM-6701 F). The melting temperatures and solid-state transitions were determined using a differential scanning calorimetry (DSC) (Metler Toledo DSC 821E) using about 5–8 mg of samples heated between 25°C and 350°C, with heating rate of 10°C/min. Nitrogen gas at a flow rate of 10 mL/h was used as the carrier gas during the measurement. The software used to analyse the output from the DSC was STARe Default DB V9.20. FTIR spectroscopy (Perkin Elmer Spectrum One) was used to study any changes in the functional group of the particles using the frequency range of 4000–500 cm\(^{-1}\). The X-ray powder diffraction (XRD) patterns were determined (Rigaku, Tokyo, Japan) with Cu target and Ni filter at 40 kV, 40 mA, scanning angle of 3–35°, and scanning speed of 1°/min.

Transmission emission microscopy (TEM) (Tecnai G2 20) analysis was used to examine the morphology of the particles whereby the encapsulated MA-βCD was prepared in two different solvents: acetone as a hydrophobic solvent and distilled water as a hydrophilic solvent. In comparison, two solutions before mixing and forming a suspension of MA-βCD were also tested, i.e., as-received MA was prepared in ethanol and βCD was prepared in distilled water. Then, two drops of each solution were placed on the TEM sample holder and the dropped solution was dried at room temperature for 10 min.

The dissolution tests for the three prepared samples, i.e., as-received MA, encapsulated MA-βCD, and unencapsulated MA particles, were determined using United States Pharmacopeia dissolution apparatus XXIV-Type II (paddle-37°C) (Electro Lab, Mumbai, India). The dissolution medium was a 900 mL phosphate buffer with pH 7.4. The amount of dissolved drug was determined using UV spectrophotometric method (UV 1601 A, Shimadzu, Japan) at 260 nm.

3. Results and discussion

3.1 Electrical properties of MA-βCD suspension in water

The stability of suspension was measured by identifying the electrical properties or zeta potential value of the dispersion. High zeta potential value indicates the solid body will remain suspended, i.e., colloidal suspension before atomization of
droplets. To obtain the highest zeta potential value, the pH of the dispersion was varied from acidic to basic condition. In this work, βCD in water and MA in ethanol represented the solutions prior to the preparation of the encapsulation mixture (colloid suspension) of MA-βCD at different pH (see Figure 3a). The zeta potential values of all samples were measured between pH 2–11, and the values differed probably due to the attachment of hydrophobic group of MA on the hydrophobic ring of βCD in the prepared suspension (after mixing). βCD in aqueous and MA-βCD suspension showed its isoelectric point at pH 2. The isoelectric point of MA in ethanol is at pH 6. MA-βCD suspension in the region of isoelectric point or having a zeta value close to these isoelectric points needs to be avoided because at isoelectric point where the zeta value is equivalent to 0 mV, the solid body in the dispersion is less stable and has high tendency to agglomerate and sediment. This phenomenon is known as specific electrostatic interaction effect (Chi et al. 2003), whereby the decrease of zeta potential leads to the decrease of electrostatic repulsion and causes aggregation (Kharia, Singhai, and Verma 2012). MA-βCD suspension showed the zeta potential magnitude value of 0.2 ± 6 mV at pH 2, which decreased to −13 ± 5 mV at pH 11. The zeta potential value for MA-βCD suspension obtained in this work was moderately low since it is difficult to obtain high zeta potential value of hydrophobic drugs in aqueous solution. This is common for hydrophobic drugs such as silymarin, β-carotene, and butylated hydroxy toluene, whereby the drugs have low zeta potential values of 3.3, −7.0, and −7.8 mV, respectively (Guo et al. 2013).

The appearance of MA-βCD suspensions at different pH is shown in Figure 3b. The observations of initial suspensions at pH 6 indicated that the particles were readily suspended, which is when the zeta value approaches the isoelectric point, and as the pH value increased, it became more stable and the particle agglomeration was reduced as at this stage the zeta value was the highest. When some NaOH was titrated to the mixture, it became more stable as the pH value increased. By adding NaOH, the agglomerated and deposited particles were reduced because the hydrogen ions released from the agglomerated surface react with hydroxyl ions from the environment. Based on this observation, the most stable region for the suspension was at pH 11. Therefore, the electrospraying of suspension for producing encapsulated MA-βCD particles was prepared at pH 11 by considering the suspension stability. At this stage, encapsulated MA-βCD is in wet form, and after the electrospraying process its become complex form of encapsulation MA-βCD particles.

The conductivity value for the suspension of encapsulated MA-βCD particles was found to be 440 μS/cm, and for the two solutions of unencapsulated MA, the conductivity values were 277 and 855 μS/cm, which can be considered large enough to form fine droplets via electrospray as studied by Lopez-Herrera et al. (2003) using ethylene glycol.

### 3.2 Morphology and size distribution of the deposited particles

Figure 4 shows the morphology of the deposited particles collected on the substrate sprayed from the electrospray. The encapsulated MA-βCD particles were nearly monodispersed with almost spherical shape (see the enlarged image in Figure 4c) and has similar morphology as the as-received MA (Figure 6a). The shape of the encapsulated MA-βCD particles was found to be in different form from the electrosprayed MA in ethanol and electrosprayed βCD from aqueous solution as shown in Figure 4a and b, respectively.

The FESEM images for the deposited encapsulated MA-βCD particles on the substrate with different WDs are shown in Figure 5. For the production of encapsulated MA-βCD particles, the substrate was positioned at a WD of 15, 20, and 25 cm from the needle tip. The result shows that the size of the encapsulated MA-βCD particles on the substrate decreases as the WD increases, and in this case, the encapsulated particles of MA-βCD reach the particle size within the nano range. For the WD of 15 and 20 cm, the deposited encapsulated MA-βCD particles are with an average size of 91 ± 26 nm and 67 ± 46 nm, respectively. The average size of the encapsulated MA-βCD particles reduces up to 42 ± 35 nm when the WD of 25 cm was used. Figure 6 shows the FESEM images of the as-received MA and unencapsulated MA at different concentrations. The unencapsulated MA solution with the concentration of 3.33 mg/mL (see Figure 6b) produces monodisperse irregular hexagonal-shape particles. The result proves the fact that the electrospray method is capable of producing monodispersed and less nonagglomerated fine particles with the right concentration compared to Figure 6c. Figure 6c shows the deposited particles of unencapsulated MA solution with the concentration of 2 mg/mL. The image shows the existence of solvent residues that might consist of impurities and incomplete mefenamic crystal formation, which indicates incomplete solvent evaporation occurred during the process. This process produces inhomogeneous semi-solid flat particles that solidify after deposition.

---

**Figure 3.** (a) Zeta potential value of MA-βCD suspension, β-CD in aqueous solution, and MA in ethanol as a function of pH. (b) The comparisons of the appearance of MA-BCD suspension at different pH.
Low solution concentration results in slow supersaturation process during the travel from the needle point to the substrate, and hence produces particle–solvent solidification and stick to each other (Bagheri-Tar, Sahimi, and Tsotsis 2007). The unencapsulated MA particles were produced from the MA solution in acetone, which is a volatile solvent. Electrospraying high concentration of MA in a volatile solvent results in the formation of larger particles compared to the encapsulated MA-βCD particles, which was sprayed from water-ethanol suspension (less volatile solvent). A more volatile solvent evaporates faster and results in rapid particle drying (Scholten et al. 2011; Bohr et al. 2012). Comparison between the size of particles of the as-received MA (see Figure 6a) and the unencapsulated MA particles shows that the unencapsulated MA produces smaller particles, which proves that the electrospray process has the ability to reduce the size of particles approximately from 500 to 1 µm. In the electrospray system, the concentration of feed solutions affects the size of particles deposited on the substrate, in which low feed concentration produces smaller particle size than high feed concentration due to the less amount of solute to be crystallized (Ambrus et al. 2013). Hence, the number of fission increases and leads to small particle size.

3.3 Prediction of primary droplet and number of fission of encapsulated MA-βCD particles

The estimation of primary droplet size was calculated using scaling laws [Equations (1)–(3)]. The experimental and theoretical number of fissions can be estimated at different distances based on the average particle size shown in the FESEM images and the predicted particle size in Equation (4), respectively. Table 2 shows the experimental and theoretical number of fissions, in which the number of fission increases with increasing of WDs. The number of fission from the experiment is in agreement with the theoretical number of fission calculated using the Ganan-Calvo model [Equation (1)], whereby the value at the furthest distance is the closest to the experimental results that were derived for the conductivity solution Ganan-calvo (1999).

3.4 Characterization of encapsulated MA-βCD and unencapsulated MA particles

In this work, DSC was used to obtain the thermal properties of the as-received materials and the particles produced from the electrospray system. Figure 7 shows the DSC heat flow of the as-received MA, as-received βCD, encapsulated particles of MA-βCD, and unencapsulated MA at concentrations of 3.33 and 2 mg/mL. The DSC profile of the as-received MA in
Table 2. Predicted primary droplet size, $d_p$, calculated using scaling laws for the encapsulated MA-βCD particle, and the theoretical and experimental number of fission occurred at different working distances.

<table>
<thead>
<tr>
<th>Model</th>
<th>Primary droplet size, $d_p$ (nm)</th>
<th>Theoretical particles from Equation (4) (nm)</th>
<th>Theoretical number of fission from Equation (7)</th>
<th>Experimental number of fission at 15 cm ($d_p = 91$ nm)</th>
<th>Experimental number of fission at 20 cm ($d_p = 67$ nm)</th>
<th>Experimental number of fission at 25 cm ($d_p = 42$ nm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fernandez de La Mora and Loscertales (1994)</td>
<td>7,066</td>
<td>268</td>
<td>18,328</td>
<td>468,162</td>
<td>1,172,995</td>
<td>4,761,820</td>
</tr>
<tr>
<td>Ganan-Calvo (1999)</td>
<td>2,503</td>
<td>95</td>
<td>18,289</td>
<td>20,809</td>
<td>52,138</td>
<td>211,657</td>
</tr>
<tr>
<td>Hartman et al. (2000)</td>
<td>3,083</td>
<td>117</td>
<td>18,296</td>
<td>38,886</td>
<td>97,430</td>
<td>395,523</td>
</tr>
</tbody>
</table>

$d_p$ is average $d_p$ value from FESEM image analysis (cf. Figure 5).

Figure 7a shows that the material is a form I MA, which then transformed to form II due to the presence of two endotherm peaks, i.e., at 177.47°C and a sharp peak at 230°C. This is in accordance with the findings of other researchers in which they reported that melting temperatures of MA are at 160–220°C (form I) and 229–230°C (form II) (Cesur and Gokbel 2008; Andrews et al. 2009). As for the as-received βCD (Figure 7b), the endothermic peak was found at around 110°C associated with loss of moisture (Calsavara, Zanin, and Moraes 2011). The encapsulated particles of MA-βCD in Figure 7c show a broad endotherm at 80–120°C due to loss of nonbounded water molecules (Racz et al. 2012) but no endotherm peak at 160–230°C in which the melting temperature of MA disappeared from the diagram. This result suggests that βCD had interacted with MA to form encapsulated MA-βCD complex during the electrospray process and hypothesized that, normally, when the guest molecules (drug) were encapsulated by the βCD, the melting, boiling, or sublimation temperature will change or disappear (Spamer et al. 2002). This indicates that the drug is already incorporated into the cavity of the cyclodextrin. The disappearance of drug melting point in the encapsulated particles is similar to the findings of previous researchers who also worked with βCD and other drugs but used a different technique (Cui et al. 2012; Inoue et al. 2013).

For unencapsulated MA particles with the solution concentrations of 3.33 and 2 mg/mL, small peaks at 170.88°C and 166.36°C (form I polymorph) and sharp peaks at 229.69°C and 229.63°C (form II polymorph) were obtained, respectively. The melting temperatures are slightly lower than the melting temperatures of the as-received MA and findings by other researchers (Panchagnula et al. 2004; Kato, Otsuka, and Matsuda 2006a; Seethalekshmi and Row 2012), but they fall within acceptable range for melting point of MA. The reduction in the value of the melting temperature is probably due to the small size of the unencapsulated MA particles, and this finding is consistent with those reported by other researchers for other types of drugs with small particle size (Radacsi et al. 2012; Ambrus et al. 2013).

Figure 8 shows the FTIR spectra of the as-received MA, as-received βCD, encapsulated MA-βCD particles, and unencapsulated MA particles from solution concentrations of 3.33 and 2 mg/mL. The FTIR spectroscopy was used to study the functional group and to identify any possible interaction between MA and βCD. The FTIR results for the as-received MA show peaks are 1595.21 cm$^{-1}$ for C = O (carbonyl) stretching vibration, 1642.26 cm$^{-1}$ for aromatics (C = C) stretching vibration, 2857.44 cm$^{-1}$ for C–H (alkanes) stretching vibration, and 3307.15 cm$^{-1}$ for N–H stretching vibration. The spectrum of the as-received βCD shows O–H stretching peak at 3303.24 cm$^{-1}$. For encapsulated particles of MA-βCD, the result depicts broader wavelength at 3000–3500 cm$^{-1}$ than the as-received MA and βCD. This peak suggests the overlapping of N–H and O–H stretching vibrations. Aromatic (C = C) and carbonyl (C = O) stretching vibration from MA molecules at the wavelength of 1642.26 and 1595.21 cm$^{-1}$, respectively, were also apparent in the encapsulated MA-βCD particles but at a low intensity. This suggested the possible interaction between MA and βCD molecules and hypothesized that MA was included in the cavity of βCD form encapsulation complex. This finding is in agreement with the previous work on encapsulating βCD with other drugs, whereby low intensity of carbonyl band (C = O) was obtained (Banik, Gogoi, and Saikia 2011). As expected, there are no significant differences in the FTIR spectra between the unencapsulated particles of
MA and the as-received MA. This finding signifies that the unencapsulated particles of MA remain as form I polymorph as the as-received sample.

Figure 9 shows the XRD patterns of the as-received MA, as-received βCD, encapsulated MA-βCD, and unencapsulated MA particles. For the as-received MA, the drug is a crystalline material with diffraction angles of 2θ at 6.3°, 13.73°, 21.3°, and 26.3°, and is in accordance with the results obtained by Kato, Otsuka, and Matsuda (2006b) for MA. The XRD patterns of βCD also show the crystalline 2θ diffraction peaks at 4.63°, 8.62°, 12.65°, 21.01°, and 22.36°. For encapsulated particles of MA-βCD, the XRD patterns show few similar peaks as the XRD patterns for MA and βCD, but there is also presence of new diffraction 2θ peaks at 11.30°, 17.60°, 18.18°, 20.36°, and 31.32° as shown in Figure 9c. The new peaks indicate the formation of a new solid phase, which probably referred to the peaks of encapsulated MA-βCD particles. This finding is consistent with the work of other researchers on encapsulation of drug with βCD (Inoue et al. 2013; Patil et al. 2013). Thus from this work, it can be concluded that the encapsulation of MA has been successfully achieved by electrospray.

Diffraction patterns for the unencapsulated particles of MA from different concentrations were also measured, and it was found that both particles of MA have similar diffraction patterns as the as-received MA. The diffraction peaks for MA particles from solution with concentration of 2 mg/mL are at 2θ of 6.27°, 13.70°, 21.30°, and 26.26°, whilst the unencapsulated MA particles with concentration 3.33 mg/mL were at 2θ of 6.28°, 13.73°, 21.33°, and 26.27°. The concentrations of MA in acetone prepared for the electrospray process (3.33 and 2 mg/mL) are much below the MA solubility point (30 mg/mL), in which crystallization would not occur if it is carried out using the cooling crystallization method. However, in this work, the use of electrospray has proved that crystallization of MA is possible even when the solution is undersaturated (see Figure 9d, e).

TEM was used to investigate the morphology of the encapsulated MA-βCD particles, as well as the MA and βCD particles. Figure 10a, b shows the TEM images for as-received βCD in distilled water and as-received MA in ethanol, respectively. Both as-received samples showed their original structure. The images in Figure 10c, d show encapsulated MA-βCD in water and acetone, respectively. The encapsulated MA-βCD particles in water are in their crystalline form and have almost similar morphology to the as-received βCD (see Figure 10a). However, the morphology of the encapsulated MA-βCD particles is almost spherical with smooth morphology and appeared as black dots in acetone. The encapsulated MA-βCD particles are hydrophobic in the inferior (facing the MA surface) and hydrophilic in the exterior. This complex structure has greatly modified the original physical and chemical properties of the guest molecules, thus making them unable to dissolve in the organic solvent such as acetone. This complex structure is also unable to aggregate with each other when it is dispersed in an organic solvent. Thus, the encapsulated MA-βCD particles remained as a stable suspension in a bulk solution. The diameters of the encapsulated MA-βCD particles are approximately between 90 and 100 nm, which is consistent with the average diameter observed in the FESEM image at a WD of 15 cm (see Section 3.2).

Figure 11 shows the dissolution studies of as-received MA, encapsulated MA-βCD, and unencapsulated MA particles of different concentrations, i.e., 3.33 and 2 mg/mL. It is evident that all the produced particles show a significant faster dissolution of MA than the as-received MA. The encapsulated MA-βCD particles demonstrated the largest percentage drug release at all time points followed by the unencapsulated MA particles. The dissolution of the encapsulated MA-βCD showed a maximum drug dissolution of 98.3% in 45 min. The dissolution rate of the encapsulated MA-βCD particles at 45 min was $1.34 \times 10^{-4} \text{mg mL}^{-1} \text{s}^{-1}$. As compared to the as-received MA particles, the dissolution was 89.5% with dissolution rate of 1.17 mg mL$^{-1} \text{s}^{-1}$. The enhancement of
The difference in the dissolution rate between the unencapsulated MA and the as-received MA at 2 and 3.33 mg/mL may be attributed to the small particle size of unencapsulated MA at 2 mg/mL, whereby fine particles dissolve faster than large particles.

4. Conclusion

In this study, we have presented the evidence that the electrospay process using cone-jet mode is able to produce near monodispersed, fine particles, and encapsulated MA-βCD particles. The size of the encapsulated MA-βCD particles is reduced to the average particle size of 42 ± 35 nm by increasing the WD between the needle point to the substrate. By using electrospray, the encapsulation of MA-βCD occurs by repetition of Coulomb fission and solvent evaporation that not only reduce the size of the particles but also envelope the MA with βCD before the solidification process takes place and reaches the substrate. The presence of the encapsulated MA-βCD is proven by the DSC result, which shows the disappearance of MA melting peak. The presence of the encapsulated MA-βCD particles is further confirmed by the new XRD peaks observed in the diffractogram pattern. The appearance of C = C at wavelength 1513–1642.26 cm$^{-1}$ in FTIR also supported the encapsulation between MA and βCD. The encapsulated MA-βCD particles dissolved much faster than the unencapsulated MA and the as-received MA. TEM result also shows that the encapsulated MA-βCD particles solubilized in water and suspended as spherical shapes in acetone. The spherical morphology of MA-βCD particles in the acetone was due to the interaction between hydrophilic outer surface of βCD with the organic solvent (hydrophobic).

Funding

The authors would like to express their gratitude to the Ministry of Education Malaysia, Universiti Teknologi MARA (UiTM) Shah Alam, Malaysia, and the Japanese government fund for Tokyo University of Agriculture and Technology, TUAT, in funding this work. This study was supported by the research grant of Research Acculturation Collaborative Efforts (RACE) [600-RMI/RACE 16/6/2 (6/2012)], Research Acculturation Grant Scheme (RAGS) [600-RMI/RAGS 5/3 (65/2012)], and Research Entity Initiative (REI) [600-RMI/DANA 5/3/REI (2/2013)] from UiTM and JSPS Kakenhi 26420761.

References


