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Preparation and Characterization of Mefenamic Acid Crystals with Surfactants-Part I-

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Abstract

The characteristics and dissolution of mefenamic acid (MA) crystals prepared in the presence of three different types of aqueous surfactants were studied; the surfactants used were namely, anionic sodium lauryl sulphate (SLS), cationic cetrimide and non-ionic surfactant (Tween 80). Various techniques were used for characterization including: scanning electron microscope, differential scanning calorimetry (DSC), fourier transform infrared (FTIR) spectroscopy and X-ray diffractometry (XRD). Dissolution profiles for the starting material and crystals were also constructed.

It was found that the starting material was mainly of polymorphic (I), the stable form. Recrystallization, using different types of aqueous surfactants yielded different crystal habits. The enhancement of dissolution rates of crystals prepared with the above mentioned aqueous surfactants were found to be in the order of tween 80> cetrimide> SLS. The mechanism of enhancement and the effect of crystal habit on dissolution were also discussed.

Keywords: Mefenamic acid, Crystal habit, Surfactants

Introduction

Mefenamic acid (MA) is at high dose, anti-inflammatory drug, which is effective, and safe in the treatment of rheumatoid arthritis, however, (MA) is a problematic drug in granulation, tabletting and dissolution due to its poor solubility, hydrophobicity and tendency to stick to surfaces (Adam et al., 2000).

In the literature several methods for improving solubility and dissolution of poorly water soluble drugs are described. Among these, solid dispersions (Gupta et al., 2002), complexation (Imai et al.,1991), microencapsulation, addition of wetting agents and surfactants (Chen et al., 2003), salt formation and solid-state manipulation (Nada et al.,2005), crystal engineering techniques are also used to modify the crystal properties such as habit, polymorphism and size. The properties of a drug can be greatly influenced by choosing a suitable polymorphic form, suitable crystal habit and suitable crystallization technique (Shekunov and York, 2000).

The habit or external shape of a crystal results from different rate of growth of the various faces, under certain conditions one set of faces may grow at a faster or slower rate than other producing altered shape. One method of affecting this crystal habit modification is by the

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addition of impurity into the crystallization medium. The impurity become physically or chemically adsorbed on to the growing crystal faces or edges thus impeding further growth. If present in small quantities, the impurity will be adsorbed preferentially to some faces and not others, depending upon the chemical nature of the faces producing a changed crystal habit since growth will occur more quickly in the direction of faces or edges will be least affected.

If present in large quantities, the impurity is likely to adsorb onto all faces and a habit will result that is quite different from that achieved in the presence of trace amounts (Attwood, 1988; Halebleian, 1975).

The overall shape of a growing crystal is determined by the relative rates of growth of its various faces, the slower the growth rate the larger is the face.

In general the growth rate of a surface will be controlled by a combination of structurally related factors, such as intermolecular bonds and dislocations, and external factors such as supersaturation, temperature, solvent and impurity concentration (Hatakka et al., 2002)

Structurally related compounds are common impurities in pharmaceutical components. These compounds are incorporated with varying efficiencies, and can, hence, influence the nucleation and subsequent crystal growth rate of the solute (Thompson et al., 2004).

Incorporation of different materials such as surfactants, polymers or additives during crystallization process has been known and tried in order to achieve substances with better properties (Rasenack and Muller, 2002a; Thompson et al., 2004).

Michaels and Coleville as cited by (Attwood, 1988) added small amounts of surface active agents (anionic and cationic) into the crystallization medium of Adipic acid. The rate of growth of the normal faces (in the presence of anionic surfactants) were found to be increased in the order (110)›(100) ›(001), with rate constants proportional to hydroxyl ion concentration on the faces, giving prismatic or needle shaped crystals. Cationic surfactants had quite the opposite effect, favoring the formation of plates or flakes. Extent of modification dependent upon concentration of impurity and also its carbon chain length

The molecular structure of a face will determine the availability of sites that favor adsorption of additives. The availability of sites and their energy on a particular crystal face will determine the extent of adsorption. The growth rate of a crystal face is decreased by adsorption of the additive (Lechuga et al., 1993).

The aims of this study includes preparation and characterization of MA crystals prepared in the presence of different types and concentrations of surfactants using different techniques such as scanning electron microscope (SEM), DSC, FTIR, and X-ray diffraction. The dissolution profiles of these crystals were also studied.

Material and Method

Mefenamic acid (MA) was supplied by JPM (Naour, Jordan), sodium lauryl sulfate (SLS), cetrimide(cetyl trimethyl ammonium bromide) , tween 80 , absolute ethanol, potassium dihydrogen orthophosphate, sodium hydroxide ,were all of pharmaceutical grade.

1. Preparation of the crystals

Different crystal samples of mefenamic acid were prepared in the presence of surfactants aiming to achieve better physico-chemical properties.

One gram of MA was dissolved in 50 ml of absolute ethanol by placing it in a steam bath; the solution was stirred continuously until it became clear. In another conical flask one gram or two grams of a surfactant was added to 50 ml of distilled water and placed on the steam bath at the same temperature. Then the aqueous surfactant solution was added to MA solution (to get a final concentration of 1% or 2% of a surfactant), stirred by glass rod for 30 seconds, labeled and left overnight at room temperature. The crystals were filtered and left on a filter paper for 24 hours in a Pyrex desiccators containing fresh silica gel. The crystals were filled into clean vials, closed, labeled then covered again with parafilm and stored in the Pyrex desiccators to be used for further studies. The same procedure was carried out to prepare MA crystals without surfactants.

2. Dissolution studies

100 mg of MA starting material and other crystallized samples were filled in a hard gelatin capsule of size 5, placed in 500ml phosphate buffer (pH 7.4) using Ereweka dissolution apparatus at a paddle speed of 100rpm with temperature maintained at 37°±1°C. Aliquots of 5 ml were withdrawn at appropriate intervals, filtered using 0.45μm millipore filter and an equal volume of phosphate buffer was replaced. MA in the aliquot was assayed spectrophotometrically at $\lambda = 333$ nm.

3. Fourier transform infrared (FTIR)

The FTIR spectroscopy was performed using a Nicolet Avatar 5.1 ESP 360 Spectrometer, connected to Omnic software run using Windows 95. The blank used was KBr; each sample was diluted by mixing of 0.1 g of the sample with 1.9 g of KBr, and then placed for analysis as powder without compression. The samples were analyzed using diffuse reflectance cells.

4. X-ray powder diffraction

Powder diffraction patterns of all samples were determined using a PW 3040 diffratometer (Xpert MPD, Phillips, Netherlands) with cobalt radiation. The sample tubes were filled completely with mefenamic acid starting material and other crystallized samples and then measured at a generator tension of 40kv and a generator current of 40 mA.

5. Differential scanning calorimetry (DSC)

Differential scanning calorimetric measurements were performed using Quick cooling differential scanning calorimeter DSC-50 Q Shimadzu, Japan, equipped with a Shimadzu TA-50 WSI instrument controller and a Shimadzu professional computer.

Samples were weighed in aluminum pans that were sealed with a crimper. The thermal behavior was studied under a dry nitrogen purge (20ml/min) at a heating rate of 10°C/min.

6. Scanning electron microscopy (SEM)

Samples were prepared by coating them with gold using a vacuum coater (Polaron E 6100 vacuum coater, UK), and examined by scanning electron microscope (Camera SU 30, Semprobe, France).

Results and Discussion

Characterization of the crystals

1. Differential scanning calorimetry

It is known that mefenamic acid has two polymorphic forms, the stable polymorph form I and the metastable polymorph (form II). Differentiation between these two polymorph forms was studied using DSC technique (Panchagnula et al., 2004 and Gilpin and Zhou, 2005).

It was found the metastable form II showed only one endothermic peak corresponding to the melting point at about 230°C. While the stable form I displayed two endothermic peaks, the first peak corresponds to the transition of form I to form II which is affected by the experimental conditions such as the heating rate. This peak was found to be at 179°C (heating rate at 40°C/min), 190°C (5°C/ min) or between 215-220°C (2 °C/ min).The second peak corresponds to the fusion of form II, it was found to be at about 230°C. Figure 1 (a - e) shows the thermograms of the starting powder of MA, MA crystals and MA crystals in presence of surfactants (cetrimide, SLS and tween 80), all of the DSC thermograms of MA show two endothermic peaks that correspond to the stable polymorph I. The first peak lies between 169-184°C, and the second peak between 226.56-228.18°C.These variations might be due to the presence of surfactants and due to crystallization conditions.

2. X- Ray Powder diffraction

Figure 2 (a-e) shows the x-ray diffraction pattern for both mefenamic acid MA starting material and mefenamic acid in the crystal forms. Samples showed similar x-ray patterns that were consistent with x-ray pattern of polymorph I. Polymorph I has a characteristic peak at $2\Theta=7°$, while presence of peak at $2\Theta=18°$ is characteristic for polymorph II (Romero et al., 1999).

Furthermore, the relative intensity of peaks varied in all samples. These variations were attributed to the differences in crystal habit and size that resulted in preferred orientation of the crystals in the x-ray diffraction sample holder (Panchagnula et al, 2004).

The x- ray diffraction pattern of the powder form shows both peaks with comparable intensities, indicating the presence of a mixture of both polymorphs.

3. Fourier transform infra red spectroscopy

The IR absorption spectra of forms I and II mefenamic acid show characteristic differences in the detailed shape and intensities of some of the major absorption bands that can be used to identify each polymorph. Specifically, in the region of wave number between 3350 cm^{-1} and 3300cm^{-1}, the N-H stretching frequency occurs at (3310-3250) cm^{-1} for form I and at 3347 cm^{-1} for form II. In plane deformation of N-H in the region (1650-1600) cm^{-1} that overlapped with the stretching band at 1650 cm^{-1}.In addition distinct absorption peaks between 1600-1060 characteristic of C-H vibration were also observed. The FTIR absorption profiles of MA in the powder and in the crystalline form are consistent with those indicating polymorph I, Figures 3 - 4 (Panchagnula et al., 2004; Gilpin and Zhou, 2005).

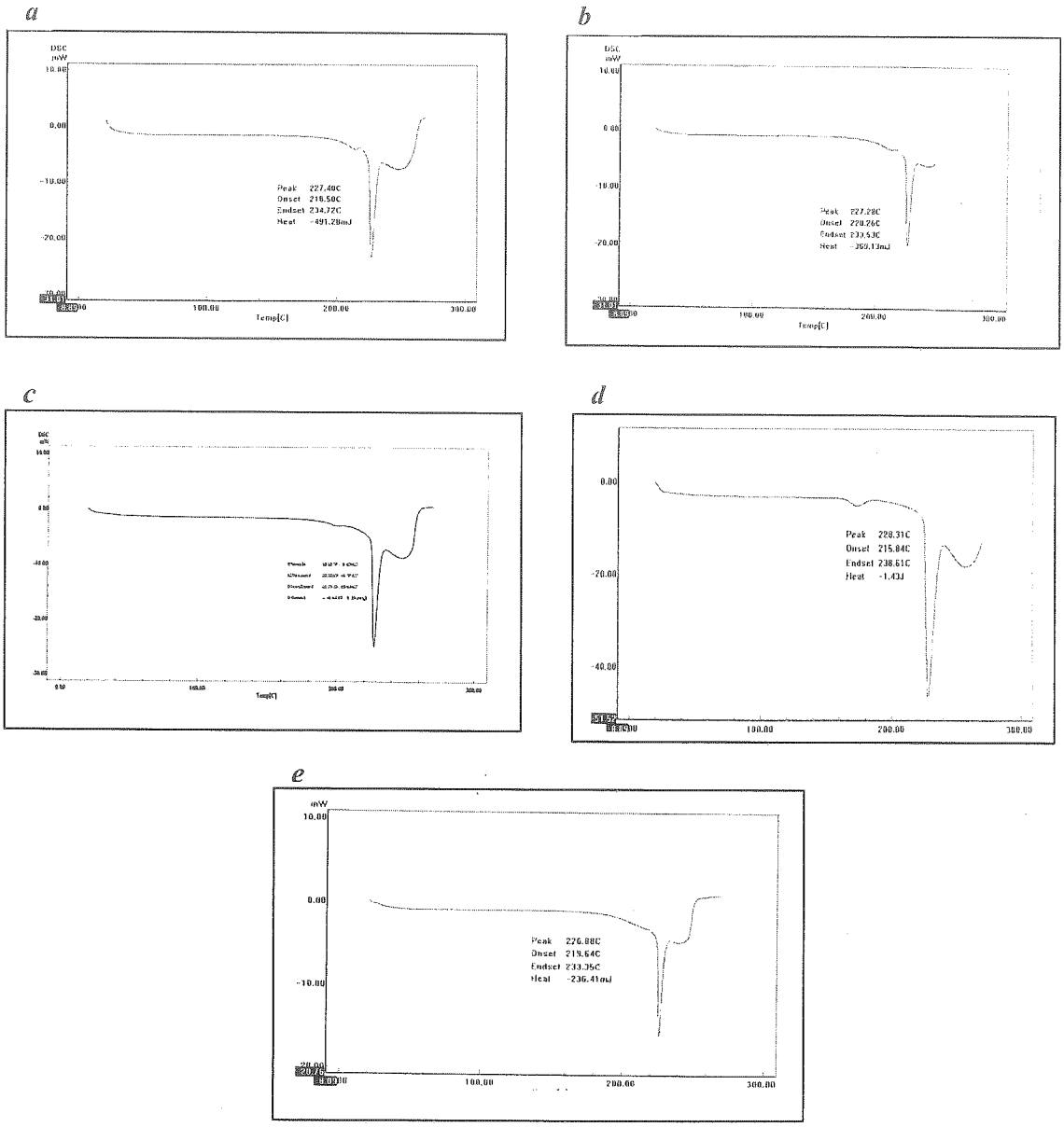


Figure 1. DSC thermograms of crystallized mefenamic acid in the presence of 1% a- Tween 80 b-cetrimide c- SLS d- mefenamic acid crystals without surfactants e- mefenamic acid powder starting material

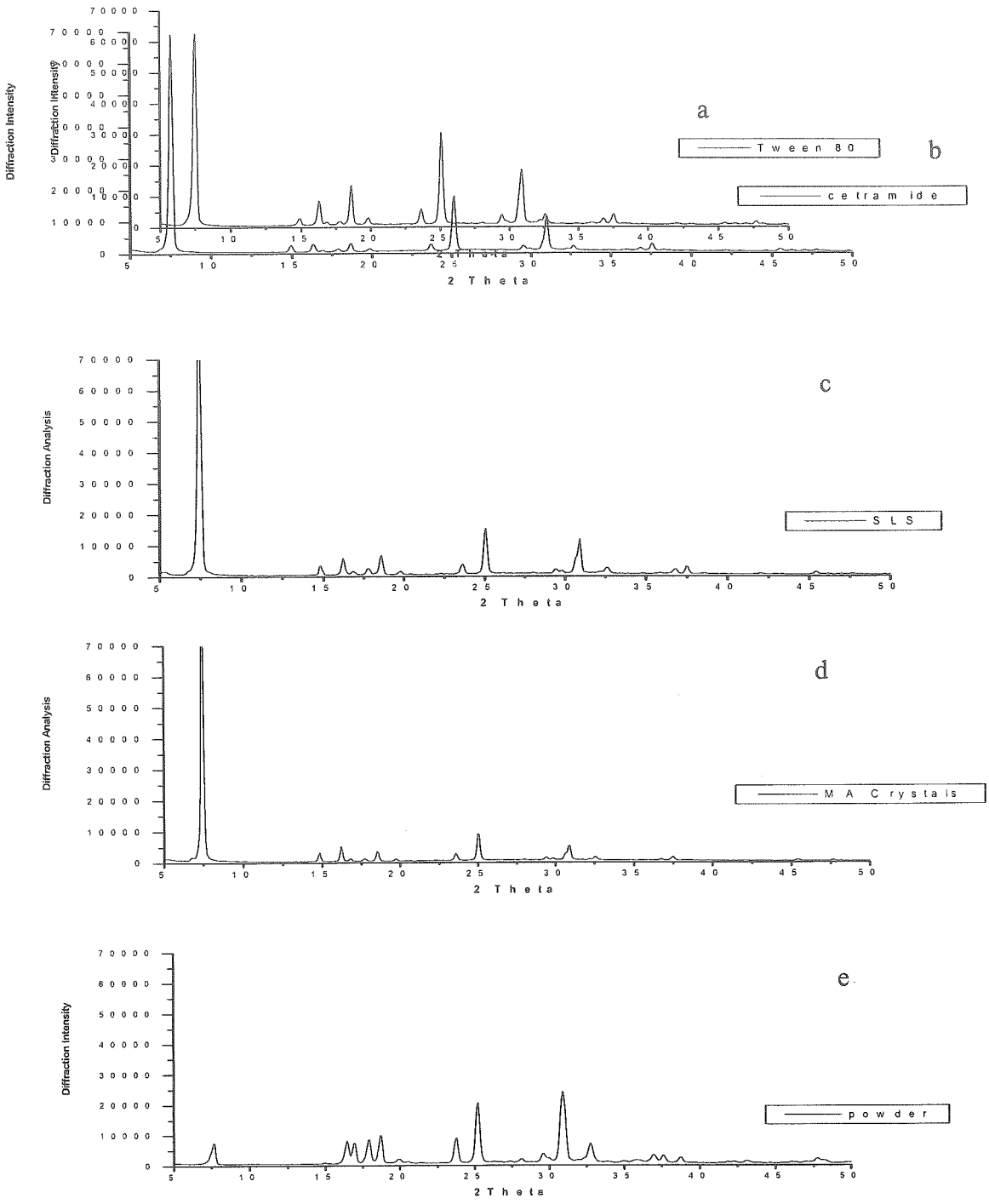
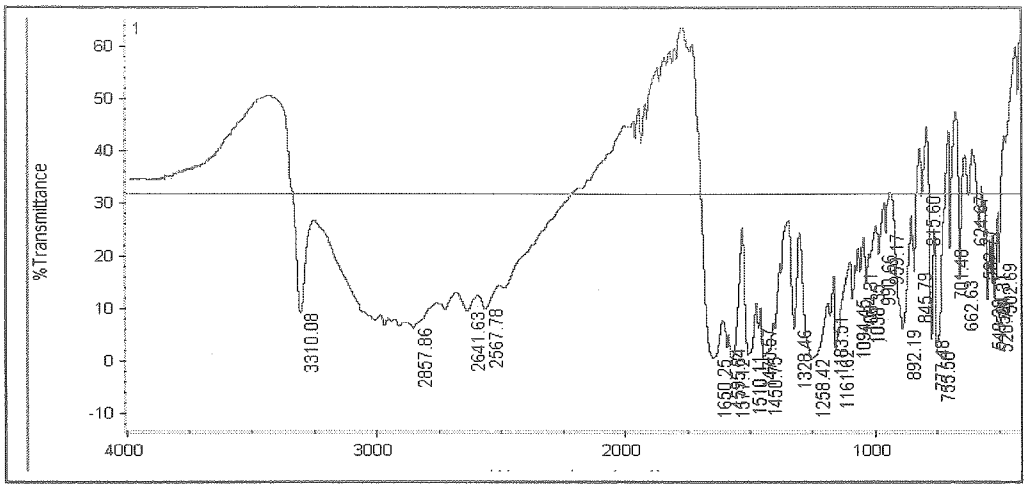
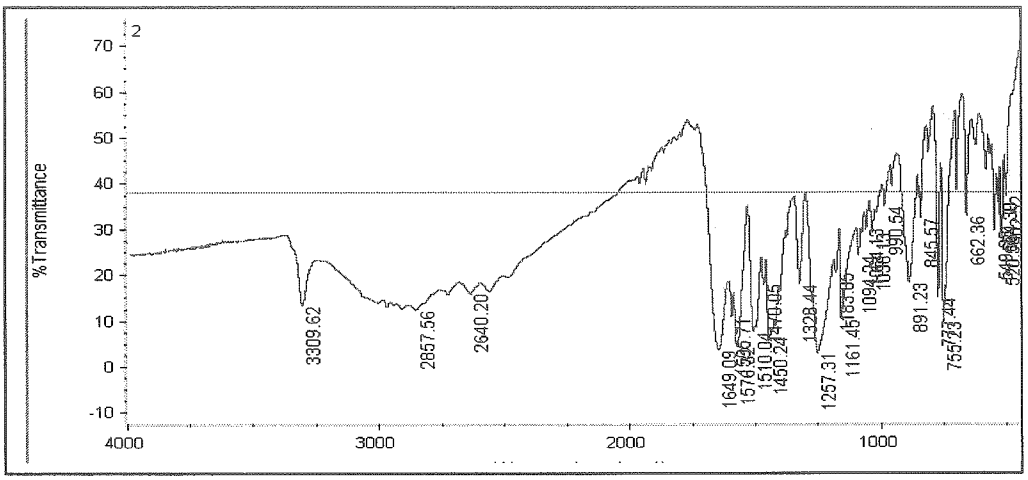


Figure 2. X-ray diffraction patterns of mefenamic acid crystallized in the presence of 1% a- cetrimide b- tween 80 c- SLS d- crystals of mefenamic acid without surfactant e- starting powder of mefenamic acid.

a-



b-



c-

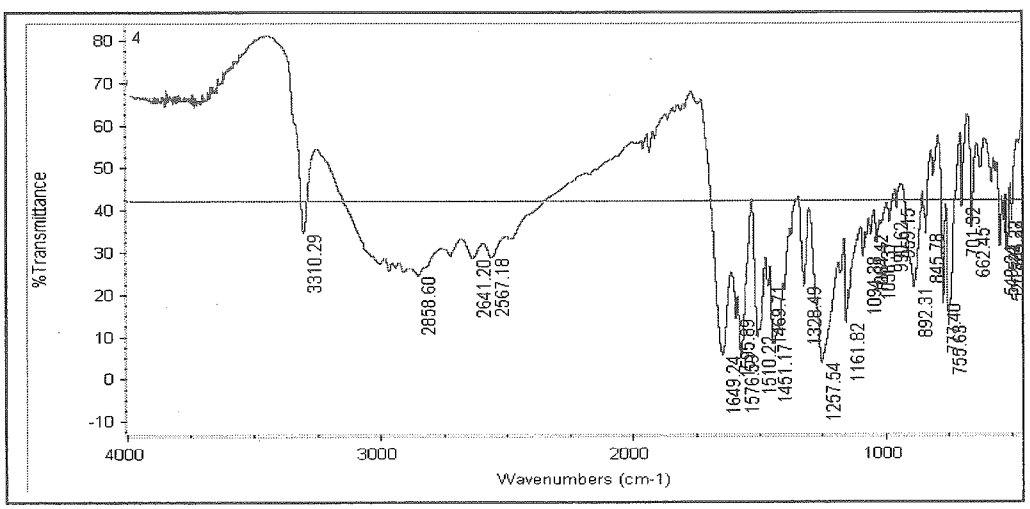
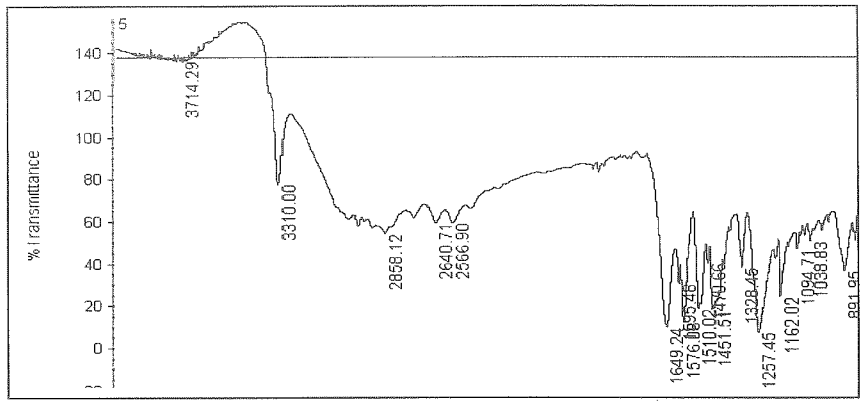


Figure 3. FTIR spectrum of mefenamic acid crystallized in the presence of 1% a- cetrimide b- Tween 80 c- SLS

a-



b-

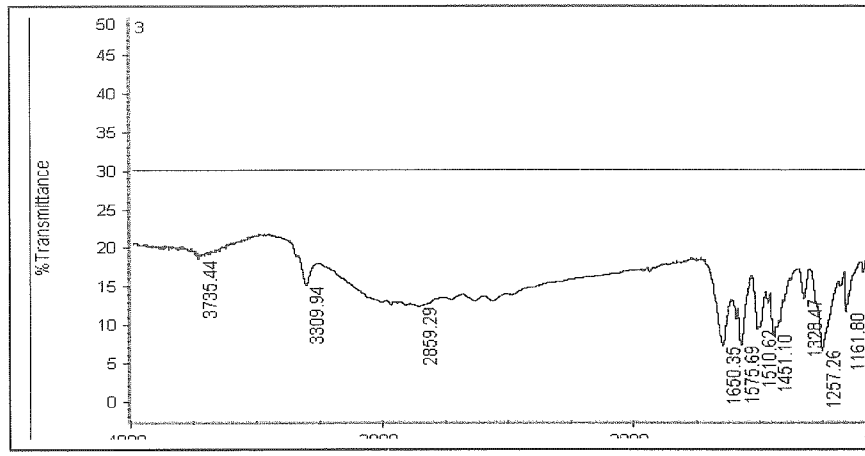


Figure 4. FTIR spectrum of mefenamic acid a- in the crystal form b- starting material

3.4. Scanning electron microscope:

SEM clearly demonstrates the difference in the crystalline form between (MA) starting material, MA crystals and MA crystallized with different surface-active agents as shown in Fig. 5 (a-e). The crystals of (MA) were predominantly hexagonal plates with smooth faces and sharp edges. Samples of crystallized (MA) in presence of SLS (anionic) were acicular, due to elongation along the c axis (in the {001} direction) and b axis (in the {110} direction) respectively. Crystals of (MA) in the presence of cetrimide (cationic) were plate shaped with angular edges; additional small particles are adhered onto larger crystals. These results were in consistent with those found by Michaels and Colville as cited by Attwood (1988).

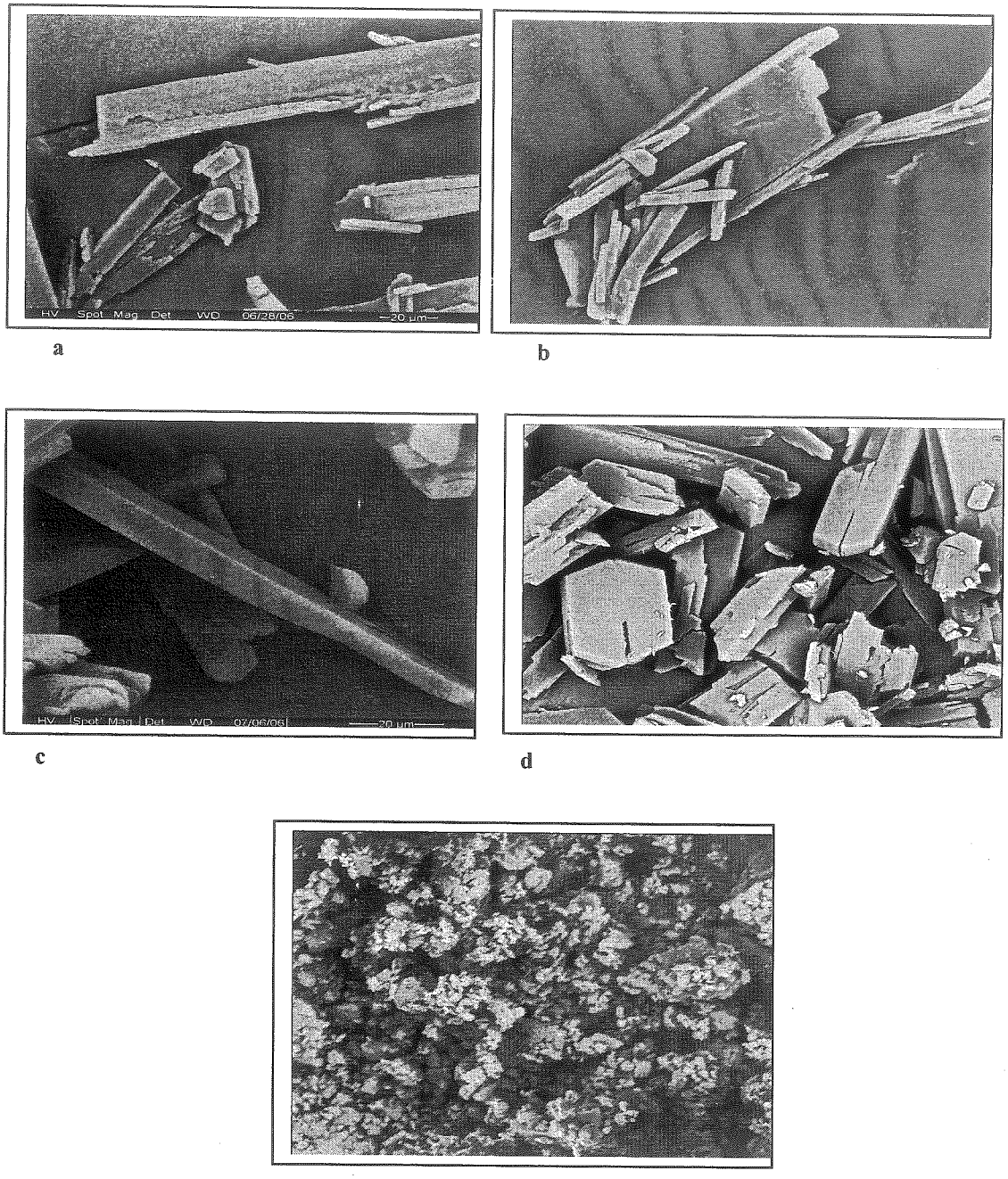


Figure 5. SEM of crystallized mefenamic acid in presence of 1% a- cetrimide b- tween 80 c- SLS d- mefenamic acid crystal without surfactant e- starting mefenamic acid powder.

The results derived from DSC, X-RAY and FTIR demonstrated that all the MA crystals studied were isomorphic. On the other hand the presence of surfactants during crystallization process of (MA) significantly affected the crystal habit of the drug.

3.5. Dissolution studies

Figure 6 illustrates the dissolution profiles of mefenamic acid MA starting material and other crystallized samples with and without surfactants in phosphate buffer at pH 7.4.

It is obvious from the figure that recrystallization from different SAA improved the dissolution rate of the drug to variable extents compared to the starting (MA) drug powder. Only 20% of the drug in the powder form was obtained after 2 hours compared with around 50% of dissolution was obtained for the crystal form of mefenamic acid. This enhancement may be due to the effect of solvent used during crystallization process (absolute ethanol), which causes hydrophillization of (MA) surface, increases its wettability and enhances the dissolution.

By comparing the dissolution of (MA) samples crystallized in the presence of surfactants with (MA) starting powder it was found that the amount released from all crystals prepared in the presence of surfactants was better than (MA) crystals or (MA) starting powder. Two factors are working at the same time to improve dissolution of MA crystals, firstly; the wettability power of the surfactant and secondly; the habit and the physical properties of the crystals. Chiou et al., (1976) postulated that surfactant molecules due to their surface activity might be adsorbed on to the hydrophobic surface of the crystals; this would increase the wettability of the crystals, and thereby increase their dissolution rate.

It was found from the above figure that the type of surfactant had a profound effect on the release of (MA) from different crystals. The crystals prepared in the presence of tween 80 (nonionic surfactant) showed the best release followed by crystals prepared in the presence of cetrimide (cationic surfactant) followed by crystals prepared in the presence of SLS (anionic surfactant). Although SLS has the highest HLB (around 40) the crystals prepared in the presence of SLS showed the lowest release. This could be attributed to several factors, among these is the effect of surfactant on the crystal habit of MA, producing acicular shape or needle shape with more cohesive property between particles and consequently less release. Another factor is the decrease in the adsorption of SLS on the surfaces of MA crystals due to the electrostatic repulsion between the ionized carboxyl groups of MA and SLS, taking in consideration that the pKa of MA is 4 and the pH of the crystallization medium is around 4.5, it is expected that half of the MA molecules are in the ionized form.

In the case of cetrimide, the attractive forces between amine groups and carboxyl groups enhanced the adsorption of cetrimide on the MA crystals leading to a better wettability and release. Also the plate shape of the crystals might have a positive impact on the release. Tween 80 being a nonionic surfactant has the ability to cover all the surfaces of MA crystals without any preference to certain sites or surfaces as in the case of cetrimide and SLS.

An increase in the concentration of surfactants from 1% to 2% led to further improvement in the dissolution rate as illustrated in figure (7), this was expected since more concentration of the surfactant potentiates the wettability and the dissolution.

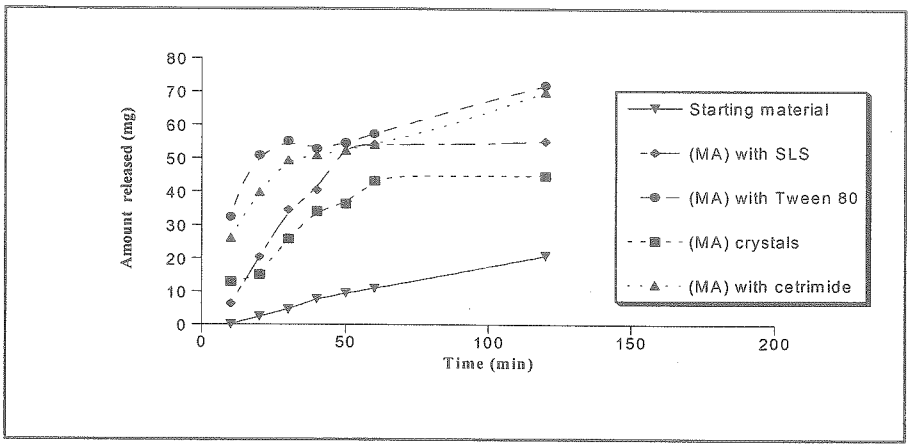


Figure 6. Dissolution profiles of mefenamic acid starting material, mefenamic acid (MA) crystals and crystallized mefenamic acid (MA) in the presence of surfactants

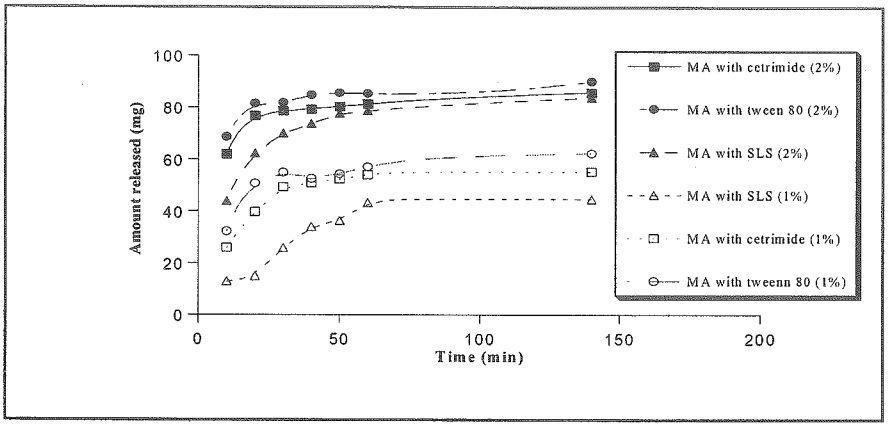


Figure 7. Dissolution profiles of crystallized mefenamic acid in the presence of different types and concentrations of surfactants.

Conclusions

Crystallization of mefenemic acid in the presence of different types of surfactants (anionic, cationic and non-ionic) led to different crystal habits of the drug depending on the type of surfactant used; these habits were confirmed using SEM. In addition to that DSC, FTIR, and X-ray were used to rule out any polymorphic changes.

Enhancement of dissolution rates due to crystallization of the drug in the presence of surfactants were in the order of tween 80 > cetrimide > SLS.

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References

Adam, A., Schrimp, L., Schimidt, P. C. (2000). Some physicochemical properties of mefenamic acid. *Drug. Dev. Ind. Pharm.* 26: 477-487.

Attwood, D. (1988). Properties of the solid state. in "Physicochemical Principles of Pharmacy" chapter 2, 2nd Ed., Macmillan press.

Chen, L.R., Wesley, J.A., Bhattachar, S., Ruiz, B., Bahash, K., and Babu, S.R. (2003). Dissolution behavior of a poorly soluble compound in the presence of Tween 80. *Pharm. Res.* 20: 797-801.

Chiou, W.L., Chen, S.J. and Athanikar, N. (1976). Enhancement of dissolution rates of poorly water-soluble drugs by crystallization in aqueous sufacatant solutions I: Sulfathiazole, prednisone and chloramphenicol. *J. Pharm.Sci.* 65: 1702-4.

Gilpin, R.K. and Zhou, W. (2005). Infrared studies of the thermal conversion of mefenamic acid between polymorphic states. *Vibrational Spectroscopy* 37: 53- 59.

Gupta, M.K., Bogner, R., Goldman, D. and Tseng, Y. (2002). Mechanism for further enhancement in drug dissolution from solid-dispersion granules upon storage. Pharm. *Dev. and Tech.* 7: 103-112.

Halebleian, J.K. (1975). Characterization of habits and crystalline modification of solids and their pharmaceutical applications. *J. Pharm. Sci.* 64: 1269-1288.

Hatakka, H., Alatalo, H. and Palosaari, S. (2002). Effect of impurities and additives on crystal growth. www2.lut.fi/ hhatakka/docit/impure. html.

Imai, T., Nohdomi, K., Acarturk, F. and Otagiri, M. (1991). Enhancement of dissolution and absorption of mefenamic acid by egg albumin. *J. Pharm. Sci.* 80: 484-487.

Lechuga-Ballesteros, D., Rodriguez-Hornedo, N. (1993). Growth and morphology of L-alanine crystals: influence of additive adsorption. *Pharm. Res.* 10: 1008-14.

Nada, A.H., Al-Saidan, S.M. and Mueller, B.W. (2005). Improving the physical and chemical properties of Ibuprofen. *Pharm.Tech.* Nov 2.

Panchagnula, R., Sundaramurthy, P., Pillai, O, Agrawal, S. (2004). Solid-state characterization of mefenamic acid. *J. Pharm. Sci.* 93: 1019-1029.

Rasenack, N. and Muller, B.M. (2002). Crystal habit and tableting behavior. *Int. J. Pharm.* 244: 45-57.

Romero, S., Escalera, B. and Bustamante, P. (1999). Solubility behavior of polymorphs I and II of mefenamic acid in solvent mixtures. *Int. J. Pharm.* 178: 193-202.

Shekunov, Y. and York, P. (2000). Crystallization process in pharmaceutical technology and drug design. *J. Crystal Growth* 211: 122-136

Thompson, C., Davies, M.C., Roberts, C.J., Tendler, S.J. and Wilkinson, M.J. (2004). The effects of additives on the growth and morphology of paracetamol (acetaminophen) crystals. *Int. J. Pharm.* 280: 137-50.

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