International Journal of Pharmaceutics 421 (2011) 230–236

Contents lists available at SciVerse ScienceDirect

International Journal of Pharmaceutics

journal homepage: www.elsevier.com/locate/ijpharm

Characterization and evaluation of miconazole salts and cocrystals for improved physicochemical properties

Shunichirou Tsutsumi[a,b,*], Motoo Iida[a], Norio Tada[a], Takashi Kojima[a], Yukihiro Ikeda[a], Toshiya Moriwaki[a], Kenjirou Higashi[b], Kunikazu Moribe[b], Keiji Yamamoto[b]

[a] Pharmaceutical Research Division, Takeda Pharmaceutical Company Ltd., 2-17-85 Jusohonmachi, Yodogawa-ku, Osaka 532-8686, Japan
[b] Graduate School of Pharmaceutical Sciences, Chiba University, 1-8-1 Inohana, Chuo-ku, Chiba 260-8675, Japan

ARTICLE INFO

Article history:
Received 10 June 2011
Received in revised form 22 August 2011
Accepted 23 September 2011
Available online 1 October 2011

Keywords:
Miconazole
Salt
Cocrystal
Crystal structure
Intrinsic dissolution rate
Stability

ABSTRACT

Miconazole salts and cocrystals were studied to improve the physicochemical properties of miconazole. Maleate, hemifumarate, and hemisuccinate were prepared and characterized by powder X-ray diffractometry, differential scanning calorimetry, and single crystal X-ray diffractometry. The intrinsic dissolution rate and stability of each miconazole crystal form were compared to those of freebase and nitrate to evaluate the optimal crystal form. Crystal structure analysis indicated that maleate was a salt formed by proton transfer from the acid to the imidazole group of miconazole. Hemifumarate and hemisuccinate were determined to be cocrystals formed by hydrogen bonding between the acids and the base in their crystal lattices. Intrinsic dissolution tests showed that the formation of salts and cocrystals improved the dissolution rate of miconazole. Stability tests of preliminary formulations prepared with each crystal form indicated that maleate and hemifumarate were unstable at 80 °C and generated a specific degraded product, i.e., a Michael adduct, between miconazole and the acids. Hemisuccinate had a superior intrinsic dissolution rate and stability, and is thus considered a promising crystal form of miconazole

1. Introduction

Crystal engineering studies are important in the pharmaceutical industry to develop stable and robust solid dosage forms of drugs, since solid-state properties have a great impact on the physicochemical and biopharmaceutical properties of drugs (Gardner et al., 2004; Huang, 2004). High-throughput crystallization screenings are commonplace in pharmaceutical research, and the technologies make it possible to obtain many types of crystal forms for drug candidates, such as polymorphs, salts, cocrystals, hydrates, and solvates (Childs et al., 2008; Kojima et al., 2006; Morissette et al., 2004).

Salt formation is a common approach to improve the solubility of poorly soluble drugs (Serajuddin, 2007). In recent years, cocrystals have also been utilized for modification of the physicochemical properties of drugs (Blagden et al., 2007; Macor, 2008). A salt is defined as a crystalline material with a solid-state assembly when proton transfer takes place from the acidic moiety to the basic moiety. On the other hand, a cocrystal is recognized as a molecular complex that contains two or more different molecules, which involve hydrogen bonding in a crystal lattice (Aakeröy et al., 2007; Childs et al., 2007). The cocrystal has gained much attention as a new solid form of drug in the pharmaceutical field, and many studies have reported the crystallization and characterization of cocrystals (Rodríguez-Hornedo et al., 2006; Schultheiss and Newman, 2009; Trask et al., 2005; Vishweshwar et al., 2006). In the drug-development process, the discovery of new crystal forms offers an opportunity to select an optimal form for a drug candidate. The most appropriate crystal form should be selected based on characterization using various analytical techniques and rational physicochemical studies that include investigations of solubility and stability (Bastin et al., 2000; Kojima et al., 2008; Limwikrant et al., 2009; Morris et al., 1994).

Miconazole (1-(2-(2,4-dichlorobenzyloxy)-2-(2,4-dichlorophenyl)ethyl)-1H-imidazole; Fig. 1a) is a broad-spectrum antifungal drug of the imidazole group, which comes in various forms, including oral, parenteral, and transdermal formulations (Nafee et al., 2003). Miconazole freebase shows very low solubility in water (less than 1 μg/mL). Many studies have attempted to improve the solubility of miconazole using cyclodextrin, surfactant, and amorphous formulations (Bhalekar et al., 2009; Jacobsena et al., 1999; Levy et al., 1995; Piel et al., 1998; Tenjarla et al., 1998). On the other hand, few studies have characterized the salts and cocrystals of miconazole. Barillaro et al. (2004) prepared maleate

* Corresponding author at: Pharmaceutical Research Division, Takeda Pharmaceutical Company Ltd., 2-17-85 Jusohonmachi, Yodogawa-ku, Osaka 532-8686, Japan. Tel.: +81 6 6300 6309; fax: +81 6 6300 6061.
E-mail address: Tsutsumi_Shunichirou@takeda.co.jp (S. Tsutsumi).

0378-5173/$ – see front matter © 2011 Elsevier B.V. All rights reserved.
doi:10.1016/j.ijpharm.2011.09.034

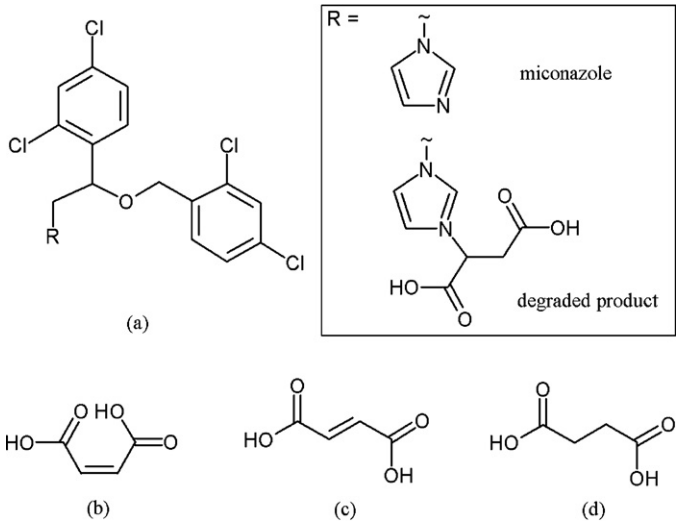

Fig. 1. Chemical structures of (a) miconazole and the degraded product, and dicarboxylic acids, (b) maleic acid, (c) fumaric acid, and (d) succinic acid.

by heating the physical mixture of its components, and Piel et al. (1998) reported the possibility of salt forms of miconazole with several acids in their solubility studies; however, these reports did not fully investigate the physicochemical properties of the miconazole salts and cocrystals. Miconazole has been developed as nitrate for a marketed form of the drug; however, nitric acid has been reported to cause ulceration and bleeding in the gastrointestinal tract (Qiu et al., 2009). For this reason, nitrate is not considered the best salt form of miconazole.

We studied the potential of salts and cocrystals of miconazole to improve its physicochemical properties by the crystallization screening, and obtained maleate, hemifumarate and hemisuccinate. This paper focuses on the physicochemical properties of maleate, hemifumarate, and hemisuccinate, as compared to free-base and nitrate. Drug–acid interactions in the solid states of these compounds were studied by single crystal X-ray diffractometry. Intrinsic dissolution tests and stability tests were performed and compared to evaluate the optimal crystal form of miconazole.

2. Materials and methods

2.1. Materials

Miconazole was purchased from LKT laboratories (St. Paul, MN, USA). Miconazole nitrate was obtained from Wako Pure Chemical Industries (Osaka, Japan). Polysorbate 80 was purchased from NOF Corporation (Tokyo, Japan). D-Mannitol was purchased from Merck (Darmstadt, Germany). All acids and solvents were obtained from Wako Pure Chemical Industries.

2.2. Sample preparation

Maleate, hemifumarate, and hemisuccinate were prepared on a 1-g scale. Precisely weighed miconazole was dissolved in 20 mL toluene at 50 °C, and 100 mM 1,4-dioxane solutions of maleic acid, fumaric acid, and succinic acid were added to prepare molar ratios of 1:1, 2:1, and 2:1 miconazole/acid mixtures, respectively. The solutions were slowly evaporated under a slow nitrogen stream at room temperature. Solids were isolated by filtration and dried under reduced pressure at room temperature. Each crystal was then gently ground in a pestle and was sieved using a 500-μm mesh to reduce particle size prior to use. The crystals for single crystal X-ray diffraction were prepared by recrystallization from a methanol solution for maleate and hemifumarate, and from toluene:1,4-dioxane (1:1, v/v) for hemisuccinate.

2.3. Powder X-ray diffraction

Powder X-ray diffraction (PXRD) patterns were collected using a Rigaku RINT 2100 Ultima IV (Rigaku, Tokyo, Japan) with Cu Kα radiation generated at 50 mA and 40 kV. A sample was placed on a silicon plate at room temperature. Data were collected from 2 to 35° (2θ) at a step size of 0.02° and scanning speed of 6°/min.

2.4. Thermal analysis

Differential scanning calorimetry (DSC) was performed using a DSC EXSTAR 6200 system (Seiko Instruments, Chiba, Japan). A DSC thermogram was obtained in a closed aluminum pan using a sample weight of ca. 3 mg and a heating rate of 5 °C/min under a nitrogen flow of 50 mL/min. Thermogravimetry (TG) was performed using a TG/DTA EXSTAR 6200 system (Seiko Instruments). A TG thermogram was obtained in an open aluminum pan under the same conditions as those for DSC at a nitrogen flow of 100 mL/min.

2.5. Single crystal X-ray diffraction

Single crystal X-ray diffraction data were recorded on a Rigaku R-AXIS RAPID (Rigaku, Tokyo, Japan) with graphite monochromated Cu-Kα radiation at 25 °C. The crystal structures were solved by direct methods and refined by a full-matrix least-squares procedure. The N–H and O–H hydrogen atoms were placed on the difference Fourier map and refined isotropically. The non-hydrogen atoms were refined anisotropically, and other hydrogen atoms were located geometrically and refined using a riding model. All calculations were performed using the Crystal Structure crystallographic software package (Rigaku) except for refinement, which was performed using SHELXL-97 (Sheldrick, 2008).

2.6. Intrinsic dissolution test

The initial dissolution rates of miconazole crystal forms were measured using a rotating disk method (Terada et al., 2000). About 20 mg drug was compressed at a force of 20 kN/cm^2 by using a single-punch tablet press (Riken Seiki, Tokyo, Japan) to obtain 7-mm diameter disks. The solvent for the dissolution test was 250 mL of 1% polysorbate 80 in the second fluid of the disintegration test, as described in the Japanese Pharmacopoeia (JP2, pH 6.8). The disk was rotated at 200 rpm and at 37 °C. At each time interval, a 0.50-mL aliquot of the solution was withdrawn from a flask and diluted to 1.0 mL with acetonitrile to provide a total dilution factor of 2. The drug concentration was determined by high performance liquid chromatography.

2.7. Stabilities of drug substances and preliminary formulations

The chemical stabilities of drug substances and preliminary formulations of each miconazole crystal form were evaluated by storing ca. 5 mg of drug substance and ca. 50 mg of preliminary formulation in closed glass vials at 4 °C and 80 °C for 4 weeks. The preliminary formulation was prepared by mixing 20 mg of each crystal form with 1980 mg D-mannitol, to a concentration of 1% (wt/wt). After storage, each sample was dissolved in water:acetonitrile (1:4, v/v) and transferred to a 25-mL volumetric flask. Preliminary formulation solutions were filtered through 0.45-μm polyvinylidene fluoride filters (Acrodisk, Pall, Port Washington, NY, USA). Drug solutions and filtrates were subjected

to high performance liquid chromatography (HPLC) to analyze chemical purity.

2.8. HPLC

HPLC was conducted using an HPLC system (W2695, Waters, Milford, MA, USA) and a UV detector (W2487, Waters) operated at 230 nm. The packaged column was Capcelpak MG III (5 μm, 4.6 × 150 mm, Shiseido, Tokyo, Japan) operated at 40 °C. The mobile phase was 50 mM sodium perchlorate buffer (pH 2.5): acetonitrile (40:60, v/v) at a flow rate of 1.0 mL/min.

2.9. Liquid chromatography–tandem mass spectrometry

Liquid chromatography–tandem mass spectrometry (LC-MS/MS) analysis was performed with an API 2000 QTRAP (Applied Biosystems/MDS Sciex, Concord, Ontario, Canada) coupled to the HPLC system, comprising an LC-10Avp (Shimadzu, Kyoto, Japan) series with degasser, autosampler, and column oven. Samples were introduced into the mass spectrometer using an Inertsil ODS-4 column (5 μm, 2.1 × 250 mm, GL Sciences, Japan) eluted at a flow rate of 0.2 mL/min at 50 °C. Elution was performed with 10 mM ammonium acetate (pH 4.0):acetonitrile (50:50, v/v). Mass spectrometric detection was performed with an API 2000 QTRAP triple quadrupole liner ion trap mass spectrometer equipped with an electrospray ionization interface in the positive ion mode. The tandem mass spectrometer was operated at unit resolution in the enhanced full mass scan mode under the following conditions: ion spray voltage, 4500 V; curtain gas, 20 psi; capillary temperature, 450 °C; declustering potential, 30 V; entrance potential, 10 V; collision energy, 30 eV and 50 eV. Data acquisition and processing were accomplished using the Applied Biosystems Analyst version 1.4.2 software.

3. Results and discussion

3.1. PXRD

The PXRD patterns of miconazole crystal forms, which were prepared in molar ratios of 1:1 for miconazole:maleic acid, 2:1 for miconazole:fumaric acid, and 2:1 for miconazole:succinic acid, were compared to those of freebase and nitrate (Fig. 2). Each crystal showed a different PXRD pattern, and the results suggested that those formed from maleic acid, fumaric acid, and succinic acid were maleate, hemifumarate, and hemisuccinate, respectively.

3.2. Thermal properties

DSC measurements were taken to investigate the thermal properties of maleate, hemifumarate and hemisuccinate, and compare them to those of freebase and nitrate. DSC curves showed different melting peaks, which were observed at 184 °C for nitrate, 140 °C for maleate, 146 °C for hemifumarate, 118 °C for hemisuccinate, and 81 °C for freebase (Fig. 3). Moreover, the formation of multiple complexes with acids increased the melting point of miconazole. TG thermograms showed that there was no weight loss until melting (Fig. 4), suggesting that the prepared crystals were not solvates. Each miconazole crystal form showed a different decomposition temperature, which was observed as a weight loss after melting, depending on their acid species.

In the DSC curves, maleate showed a unique thermal event that had an endothermic peak at 140 °C and an exothermic peak at 143 °C. To investigate whether the thermal event was derived from a polymorph transition or a chemical reaction, maleate was heated at 160 °C for 10 min and then was analyzed by HPLC. The retention time of miconazole was 8.4 min, whereas heated maleate showed a different retention time of 5.0 min, based on HPLC chromatograms (Fig. 5). The heated maleate was also analyzed by LC-MS/MS. The *m/z* value for $[M+H]^+$ was determined to be 533. The mass of this product corresponded to the sum of miconazole (molecular weight: 416) and maleic acid (molecular weight: 116), and it was considered to be a Michael adduct between miconazole and maleic acid. Barillaro et al. (2004) reported that miconazole formed into maleate after heating the physical mixture of miconazole and maleic acid, and that its melting point was 210 °C, which was actually the

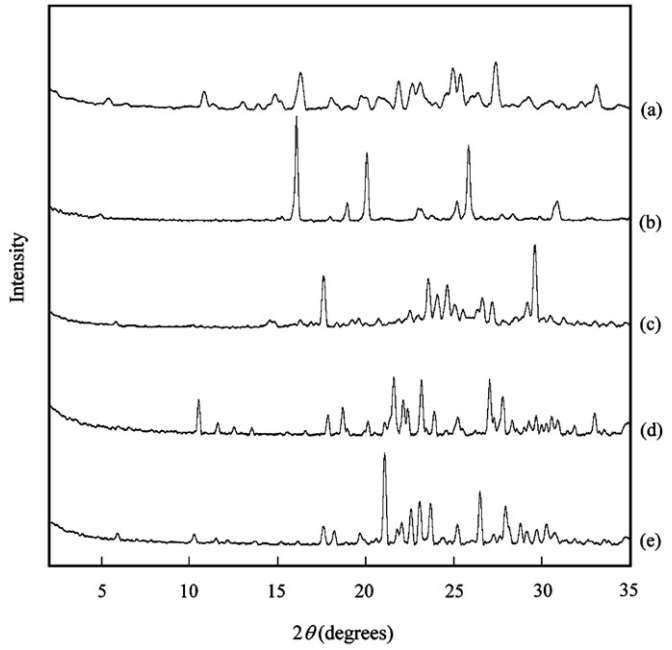

Fig. 2. PXRD patterns of miconazole crystal forms; (a) freebase, (b) nitrate, (c) maleate, (d) hemifumarate, and (e) hemisuccinate.

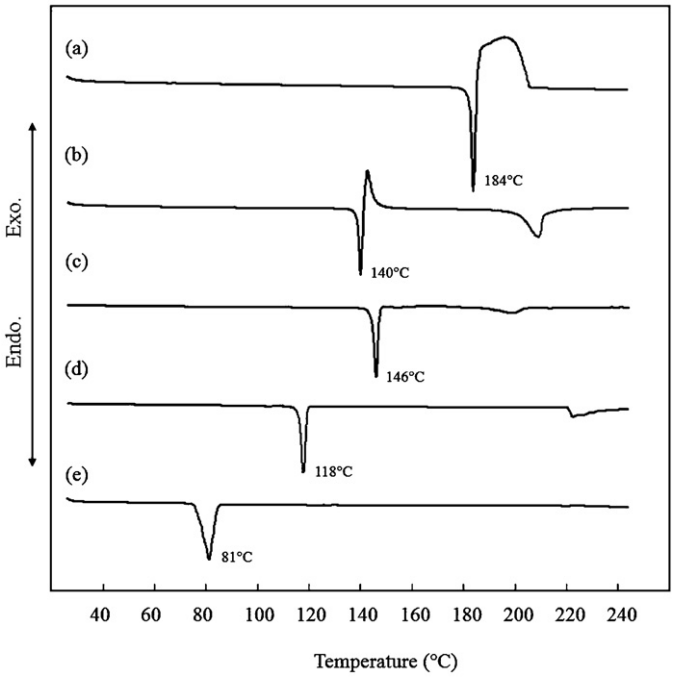

Fig. 3. DSC thermograms of miconazole crystal forms; (a) nitrate, (b) maleate, (c) hemifumarate, (d) hemisuccinate, and (e) freebase.

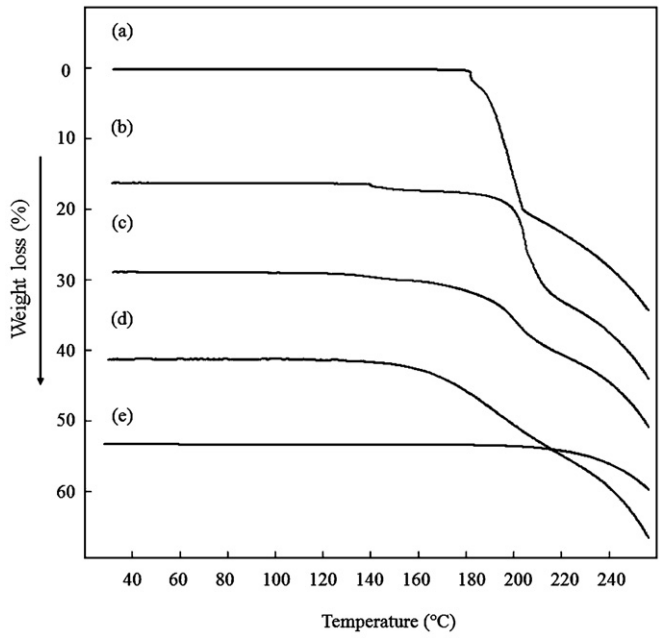

Fig. 4. TG thermograms of miconazole crystal forms; (a) nitrate, (b) maleate, (c) hemifumarate, (d) hemisuccinate, and (e) freebase.

melting point of the Michael adduct. However, this research could correct the melting point of maleate as 140 °C.

3.3. Single crystal X-ray diffraction analysis

The crystal structures of maleate, hemifumarate, and hemisuccinate were confirmed by single crystal X-ray diffractometry. The crystallographic data of these crystals are summarized in Table 1. Each crystal was triclinic and the space group was P-1. The molar ratios of miconazole and counter acids in their crystal lattices were 1:1 for miconazole:maleic acid and 2:1 for both miconazole:fumaric acid and miconazole:succinic acid (Fig. 6).

It is important to distinguish between a salt and a cocrystal for a solid-form drug composed of an acid–base complex, from both a regulatory and intellectual property perspective (Li et al., 2006; Trask, 2007). To examine the molecular states of the miconazole crystal forms, the locations of their acidic protons were investigated. In maleate, protonation was observed from one carboxylic

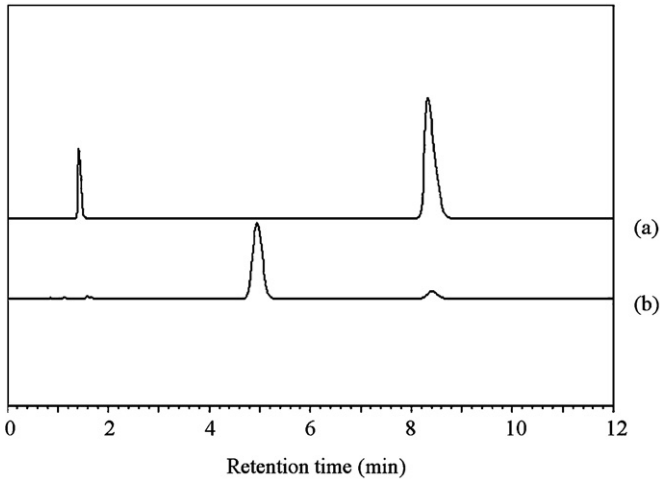

Fig. 5. HPLC chromatograms of (a) miconazole maleate and (b) miconazole maleate heated at 160 °C.

acid group of maleic acid to the imidazole group of miconazole (Fig. 6a). The other carboxylic acid group of maleic acid formed an intramolecular hydrogen bond with the carbonyl group that interacted with the imidazole group of miconazole. For hemifumarate and hemisuccinate, hydrogen bonds were observed between carboxylic acids and the imidazole group of miconazole (Fig. 6b,c).

Hydrogen positions between dicarboxylic acids and the imidazole group of miconazole were also identified by difference Fourier maps. In maleate, the N-H distance (1.070 Å) was shorter than the H to O distance (1.643 Å), thus this hydrogen atom was considered to be an acidic proton. In contrast, typical O-H bond distances were observed as 0.950 Å for hemifumarate and 0.959 Å for hemisuccinate, and the H to N distances in these crystal forms were 1.663 Å and 1.697 Å, respectively. These bond distances indicated that the protons related to hydrogen bonds between miconazole and the dicarboxylic acids. However, X-ray diffraction does not allow determining a position of the hydrogen atom with an accuracy that is sufficient for a quantitative discussion.

Several studies (Aakeröy et al., 2007; Umeda et al., 2009) have classified salts and cocrystals by the state of their carboxylic moieties, measuring the C–O and C=O bond distances determined by single crystal X-ray diffraction. In these studies, a typical C=O bond distance was around 1.2 Å and that of a C–O bond was around 1.3 Å. However, the ratio of C–O (long) to C–O (short) bonds will be very similar if deprotonation has occurred in a carboxylic acid. The ratios of C–O (long; a) to C–O (short; b) bonds in maleate, hemifumarate, and hemisuccinate are summarized in Table 2 and Fig. 6. In maleate, the ratio of C–O bond length in the carboxylic acid that interacted with the imidazole group of miconazole was 1.012 (Fig. 6a), which indicated that deprotonation occurred. Furthermore, maleic acid itself formed an intramolecular hydrogen bond, and the ratio of the bond length of the carboxylic acid was 1.065, which indicated that no deprotonation occurred. In hemifumarate and hemisuccinate, the ratios of C–O bond length were 1.075 and 1.062, respectively, which indicated that no deprotonation occurred with the carboxylic acids (Fig. 6b,c). Thus, maleate was classified as a salt, and hemifumarate and hemisuccinate were determined to be cocrystals.

The single X-ray analyses also revealed that slight differences in acid structure significantly affect the crystalline state of miconazole. Childs et al. (2007) proposed that the ΔpK_a value (pK_a of base - pK_a of acid) was an indicator of salt formation when greater than 3, cocrystal formation when less than 0, and ambiguous when between 0 and 3. Miconazole is a weak base and the pK_a value is 6.7, whereas maleic, fumaric, and succinic acids have two carboxylic groups with different backbones and their pK_a values of the first ionizable carboxylic groups are 1.9, 3.0, and 4.2, respectively. Using the ΔpK_a cutoff, maleate (ΔpK_a; 4.8) and hemifumarate (ΔpK_a; 3.7) were determined to be salts, whereas hemisuccinate (ΔpK_a; 2.5) was unpredictable, which suggests that it is difficult to predict the molecular states of miconazole crystal forms using the pK_a values of dicarboxylic acids. Although dicarboxylic acids are often used for salt and cocrystal screening for drugs, it is important to select a variety of acid species regardless of acid strength, since the molecular states of crystalline solids are determined by molecular geometry, acidity, hydrogen bond functional groups, and molecular interactions.

Single crystal analysis provided a better understanding of the solid-state interactions in the complexes of miconazole and dicarboxylic acids.

3.4. Intrinsic dissolution tests

Intrinsic dissolution tests were performed to compare the dissolution rates of maleate, hemifumarate, hemisuccinate, nitrate, and freebase. Fig. 7 shows the results in JP2 with 1% polysorbate 80.

Table 1
Crystallographic data on miconazole crystals formed with dicarboxylic acids.

	Maleate	Hemifumarate	Hemisuccinate
Empirical formula	$C_{22}H_{18}Cl_4N_2O_5$	$C_{20}H_{16}Cl_4N_2O_3$	$C_{20}H_{17}Cl_4N_2O_3$
Formula weight	532.2	474.2	475.2
Temperature (°C)	25	25	25
Crystal system	Triclinic	Triclinic	Triclinic
Space group	P-1	P-1	P-1
a (Å)	8.8476 (16)	8.2005 (10)	8.1621 (4)
b (Å)	8.9198 (16)	8.9649 (2)	9.0926 (4)
c (Å)	30.3837 (6)	15.1399 (3)	15.3327 (6)
α (°)	83.2676 (7)	82.2219 (9)	83.4506 (19)
β (°)	85.9405 (7)	81.2682 (10)	79.916 (2)
γ (°)	89.6821 (7)	70.9915 (9)	72.095 (2)
Volume (Å^3)	2375.31 (8)	1035.74 (4)	1063.86 (7)
Z	4	2	2
Calculated density (g/cm^3)	1.488	1.520	1.483
R1	0.044	0.051	0.055
wR2 (all data)	0.135	0.141	0.187

The miconazole salts and cocrystals showed approximately 2–2.5 times higher dissolution rates than freebase in 1% polysorbate 80 in JP2 at 37 °C (Table 3). These results indicate that salt and cocrystal formations of miconazole with dicarboxylic acids improve the intrinsic dissolution rate of miconazole.

Powder dissolution experiments are also important to evaluate the solubility of the miconazole crystal forms. In our preliminary powder dissolution experiments by a shake-flask method, each miconazol crystal form showed almost the same solubility (ca. 600 μg/mL) in 1% polysorbate 80 in JP2 at 37 °C for 2 h. After the solubility experiments, all miconazole salts and cocrystals were partially dissociated to freebase analyzed by PXRD (data

Table 2
The ratio of the C–O (long) to C–O (short) bond lengths of miconazole crystals formed with dicarboxylic acids.

	C–O (long; a)	C–O (short; b)	Ratio (a/b)
Maleate	1.265[a]	1.250[a]	1.012
	1.298[b]	1.219[b]	1.065
Hemifumarate	1.291	1.201	1.075
Hemisuccinate	1.275	1.201	1.062

[a] The C–O (long) and C–O (short) bond lengths of maleic acid interacted with the imidazole of miconazole.
[b] The C–O (long) and C–O (short) bond lengths of maleic acid formed an intramolecular hydrogen bond.

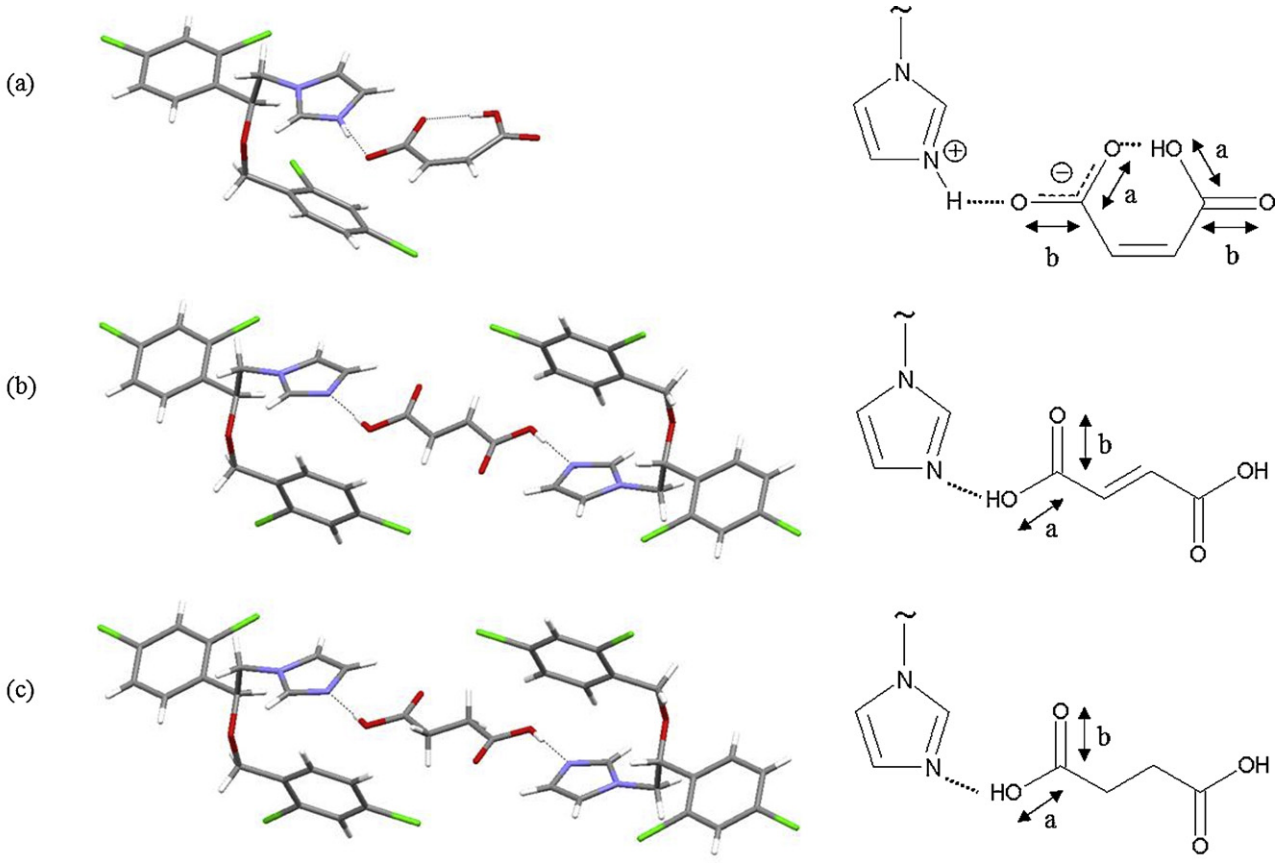

Fig. 6. Packing diagrams of miconazole crystal forms, and the interaction between the imidazole group of miconazole and dicarboxylic acids; (a) maleate, (b) hemifumarate, and (c) hemisuccinate.

Table 3
Intrinsic dissolution rates of miconazole crystal forms in 1% polysorbate 80 in JP2 at 37 °C.

	Free base	Nitrate	Maleate	Hemifumarate	Hemisuccinate
Intrinsic dissolution rate (μg/mm²/min)	0.17	0.34	0.44	0.43	0.36

Table 4
Stability of miconazole crystal forms under various storage conditions.

Sample	Storage condition	Period	Crystal form				
			Free base	Nitrate	Maleate	Hemifumarate	Hemisuccinate
Drug substance	4 °C (closed)	4W	>99%	>99%	>99%	>99%	>99%
	80 °C (closed)	4W	>99%	>99%	>99%	>99%	>99%
Preliminary formulation	4 °C (closed)	4W	>99%	>99%	>99%	>99%	>99%
	80 °C (closed)	4W	97%	>99%	75%	97%	>99%

not shown). Thus, each miconazole crystal form appeared to be physically unstable under the solution condition.

In summary, intrinsic dissolution tests were effective for the evaluation of solubility of miconazole salts and cocrystals to obtain quantitative information on the dissolution rates of miconazole crystal forms, which were not apparent from the powder dissolution experiments.

3.5. Stabilities of drug substances and preliminary formulations

The tests were performed as described in Section 2.7, and the results are summarized in Table 4. The purities of all drug substances were more than 99% at 80 °C for 4 weeks, and all crystal forms were stable in their solid states. The purities of preliminary formulations at 80 °C for 4 weeks were 97% for freebase, 75% for maleate, and 97% for hemifumarate, and their crystal forms were unstable. On the other hand, the purities of preliminary formulations of nitrate and hemisuccinate were greater than 99%, and their crystal forms were quite stable. In summary, the preliminary formulations of miconazole crystal forms varied in stability depending on their acid species, and their degradation was temperature dependent.

In preliminary formulation stability tests of maleate and hemifumarate at 80 °C for 4 weeks, specific degraded product peaks were detected at 5.0 min on HPLC chromatograms (Fig. 8), whereas the preliminary formulation of freebase generated several minor peaks of degraded products, which showed different retention times than the degraded products of maleate and hemifumarate (data not shown). Thus, the mechanisms of degradation in the preliminary formulations of miconazole crystal forms differed depending on the counter acids. LC-MS/MS analysis was performed to confirm the degraded products in the preliminary formulations of maleate and hemifumarate. The $[M+H]^+$ ion of degraded products was detected at m/z 533, which corresponded to the sum of the molecular masses of miconazole and maleic acid or fumaric acid, and its fragment ion was detected at m/z 185, which was determined to be the imidazole group of miconazole added to maleic acid or fumaric acid. LC-MS/MS analysis indicated that the degraded product was the Michael adduct of miconazole and the acids (Fig. 1); it was the same compound as that observed in the DSC study of maleate.

Some studies (Pan et al., 2011; Schildcrout et al., 1993) have reported that dicarboxylic acids that were components of salts reacted with their parent drugs and generated Michael adducts in stability tests. It has also been reported that low-dose formulations generally have poor chemical stability, because the higher the dilution ratio of excipients to drug substance, the more easily the drug substance is degraded (Badawy et al., 1999). These tendencies of drug degradation were also observed in the stability tests of preliminary formulations of miconazole crystal forms in the present study. It is noteworthy, however, that the stabilities of miconazole salts and cocrystals were different. Both maleic acid and fumaric acid have a double bond in their chemical structure, and they underwent Michael addition with miconazole. In contrast,

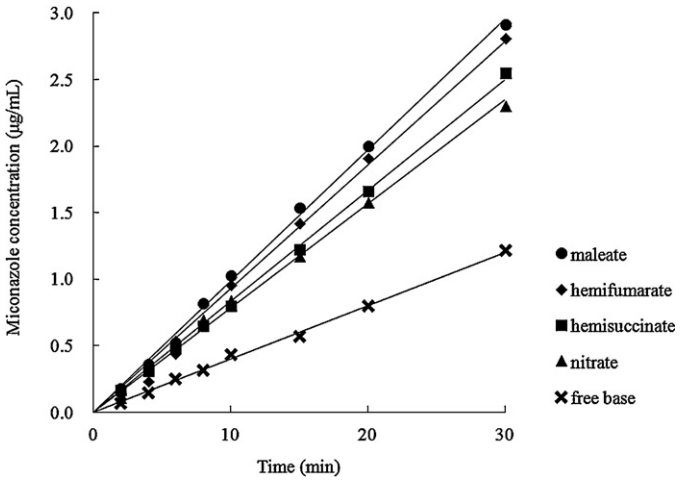

Fig. 7. Intrinsic dissolution profiles of miconazole crystal forms in 1% polysorbate 80 in JP2 at 37 °C.

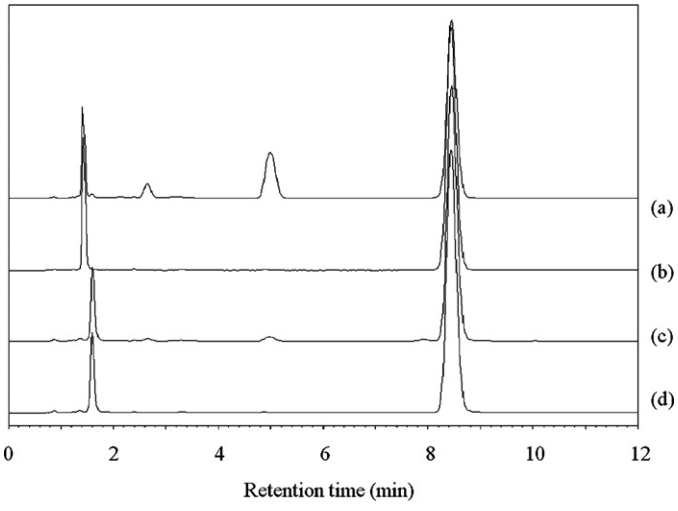

Fig. 8. HPLC chromatograms of miconazole crystal forms; (a) maleate preliminary formulation at 80 °C for 4 weeks, (b) maleate preliminary formulation, (c) hemifumarate preliminary formulation at 80 °C for 4 weeks, and (d) hemifumarate preliminary formulation.

succinic acid does not have a double bond and it did not react with miconazole.

A stability test of a low dose preliminary formulation detected a potential instability risk of a salt and cocrystal of miconazole. A crystal form of miconazole should be selected based on having a high solid-state stability. Hemisuccinate and nitrate showed reasonable stabilities under thermal stress.

4. Conclusion

Novel miconazole crystal forms (maleate, hemifumarate, and hemisuccinate) were characterized and compared to freebase and nitrate to identify an optimal crystal form of miconazole. Single X-ray diffraction analysis revealed that maleate is a salt, whereas hemisuccinate and hemifumarate are cocrystals. Intrinsic dissolution tests showed that both the salt and cocrystal forms improved solubility. Stability tests of preliminary formulations indicated that freebase, maleate, and hemifumarate were unstable at 80 °C, and Michael addition occurred between miconazole and unsaturated dicarboxylic acids. On the other hand, hemisuccinate and nitrate were quite stable in their preliminary formulations.

In conclusion, hemisuccinate is a novel cocrystal form of miconazole with an improved dissolution rate and superior stability, making it an optimal alternative to nitrate.

References

Aakeröy, C.B., Fasulo, M.E., Desper, J., 2007. Cocrystal or salt: does it really matter? Mol. Pharm. 4, 317–322.

Badawy, S.I., Williams, R.C., Gilbert, D.L., 1999. Chemical stability of an ester prodrug of a glycoprotein IIb/IIIa receptor antagonist in solid dosage forms. J. Pharm. Sci. 88, 428–433.

Barillaro, V., Bertholet, P., Henry de Hassonville, S., Ziemon, E., Evrard, B., Delattre, L., Piel, G., 2004. Effect of acidic ternary compounds on the formation of miconazole/cyclodextrin inclusion complexes by means of supercritical carbon dioxide. J. Pharm. Pharm. Sci. 7, 378–388.

Bastin, R.J., Bowker, M.J., Slater, B.J., 2000. Salt selection and optimisation procedures for pharmaceutical new chemical entities. Org. Proc. Res. Dev. 4, 427–435.

Bhalekar, M.R., Pokharkar, V., Madgulkar, A., Patil, N., Patil, N., 2009. Preparation and evaluation of miconazole nitrate-loaded solid lipid nanoparticles for topical delivery. AAPS Pharm. Sci. Tech. 10, 289–296.

Blagden, N., Dematas, M., Gavan, P., York, P., 2007. Crystal engineering of active pharmaceutical ingredients to improve solubility and dissolution rates. Adv. Drug Deliv. Rev. 59, 617–630.

Childs, S.L., Stahly, G.P., Park, A., 2007. The salt-cocrystal continuum: the influence of crystal structure on ionization state. Mol. Pharm. 4, 323–338.

Childs, S.L., Rodríguez-Hornedo, N., Reddy, L.S., Jayasankar, A., Maheshwari, C., McCausland, L., Shipplett, R., Stahly, B.C., 2008. Screening strategies based on solubility and solution composition generate pharmaceutically acceptable cocrystals of carbamazepine. Cryst. Eng. Commun. 10, 856–864.

Gardner, C.R., Walsh, C.T., Almarsson, Ö., 2004. Drugs as materials: valuing physical form in drug discovery. Nat. Rev. Drug Discov. 3, 926–934.

Huang, L., 2004. Impact of solid state properties on developability assessment of drug candidates. Adv. Drug Deliv. Rev. 56, 321–334.

Jacobsena, J., Bjerregaarda, S., Pedersen, M., 1999. Cyclodextrin inclusion complexes of antimycotics intended to act in the oral cavity - drug supersaturation, toxicity on TR146 cells and release. Eur. J. Pharm. Biopharm. 48, 217–224.

Kojima, T., Katoh, F., Matsuda, Y., Teraoka, R., Kitagawa, S., 2008. Physicochemical properties of tamoxifen hemicitrate sesquihydrate. Int. J. Pharm. 352, 146–151.

Kojima, T., Onoue, S., Murase, N., Katoh, F., Mano, T., Matsuda, Y., 2006. Crystalline form information from multiwell plate salt screening by use of Raman microscopy. Pharm. Res. 23, 806–812.

Levy, M.Y., Polacheck, I., Barenholz, Y., Benita, S., 1995. Efficacy evaluation of a novel submicron miconazole emulsion in a murine cryptococcosis model. Pharm. Res. 12, 223–230.

Li, Z.J., Abramov, Y., Bordner, J., Leonard, J., Medek, A., Trask, A.V., 2006. Solid-state acid-base interactions in complexes of heterocyclic bases with dicarboxylic acids: crystallography, hydrogen bond analysis, and ^{15}N NMR spectroscopy. J. Am. Chem. Soc. 128, 8199–8210.

Limwikrant, W., Higashi, K., Yamamoto, K., Moribe, K., 2009. Characterization of ofloxacin-oxalic acid complex by PXRD, NMR, and THz spectroscopy. Int. J. Pharm. 382, 50–55.

Macor, J.E., 2008. Annual reports in medicinal chemistry. In: Meanwell, N. (Ed.), The Emerging Utility of Co-Crystals in Drug Discovery and Development, vol. 43. Academic Press, London, pp. 373–404.

Morissette, S.L., Almarsson, Ö., Peterson, M.L., Remenar, J.F., Read, M.J., Lemmo, A.V., Ellis, S., Cima, M.J., Gardner, C.R., 2004. High-throughput crystallization: polymorphs, salts, co-crystals and solvates of pharmaceutical solids. Adv. Drug Deliv. Rev. 23, 275–300.

Morris, K.R., Fakes, M.G., Thakur, A.B., Newman, A.W., Singh, A.K., 1994. An integrated approach to the selection of optimal salt form for a new drug candidate. Int. J. Pharm. 105, 209–217.

Nafee, N.A., Ismail, F.A., Boraie, N.A., Mortada, L.M., 2003. Mucoadhesive buccal patches of miconazole nitrate: in vitro/in vivo performance and effect of ageing. Int. J. Pharm. 264, 1–14.

Pan, C., Liu, F., Motto, M., 2011. Identification of pharmaceutical impurities in formulated dosage forms. J. Pharm. Sci. 100, 1228–1259.

Piel, G., Evrard, B., Fillet, M., Llabres, G., Delattre, L., 1998. Development of a non-surfactant parenteral formulation of miconazole by the use of cyclodextrins. Int. J. Pharm. 169, 15–22.

Qiu, Y., Chen, Y., Zhang, G.G.Z., Liu, L., Porter, W.R., 2009. Developing solid oral dosage forms: pharmaceutical theory and practice. In: Tong, W.Q.T. (Ed.), Salt Screening and Selection. Macmillan, New York, pp. 75–86.

Rodríguez-Hornedo, N., Nehm, S.J., Seefeldt, K.F., Pagán-Torres, Y., Falkiewicz, C.J., 2006. Reaction crystallization of pharmaceutical molecular complexes. Mol. Pharm. 3, 362–367.

Schildcrout, S.A., Risley, D.S., Kleemann, R.L., 1993. Drug-excipient interactions of seproxetine maleate hemi-hydrate: isothermal stress methods. Drug Dev. Ind. Pharm. 19, 1113–1130.

Schultheiss, N., Newman, A., 2009. Pharmaceutical cocrystals and their physicochemical properties. Cryst. Growth Des. 9, 2950–2967.

Serajuddin, A., 2007. Salt formation to improve drug solubility. Adv. Drug Deliv. Rev. 59, 603–616.

Sheldrick, G.M., 2008. A short history of SHELX. Acta. Cryst. A64, 112–122.

Tenjarla, S., Puranajoti, P., Kasina, R., Mandal, T., 1998. Preparation, characterization, and evaluation of miconazole-cyclodextrin complexes for improved oral and topical delivery. J. Pharm. Sci. 87, 425–429.

Terada, K., Kitano, H., Yoshihashi, Y., Yonemochi, E., 2000. Quantitative correlation between initial dissolution rate and heat of solution of drug. Pharm. Res. 17, 920–924.

Trask, A.V., 2007. An overview of pharmaceutical cocrystals as intellectual property. Mol. Pharm. 4, 301–309.

Trask, A.V., Motherwell, W.D., Jones, W., 2005. Pharmaceutical cocrystallization: engineering a remedy for caffeine hydration. Cryst. Growth Des. 5, 1013–1021.

Umeda, Y., Fukami, T., Furuishi, T., Suzuki, T., Tanjoh, K., Tomono, K., 2009. Characterization of multicomponent crystal formed between indomethacin and lidocaine. Drug Dev. Ind. Pharm. 35, 843–851.

Vishweshwar, P., McMahon, J.A., Bis, J.A., Zaworotko, M.J., 2006. Pharmaceutical cocrystals. J. Pharm. Sci. 95, 499–516.