AAPS PharmSciTech, Vol. 11, No. 3, September 2010 (© 2010)
DOI: 10.1208/s12249-010-9498-5

Brief/Technical Note

Parallel Thermal Analysis Technology Using an Infrared Camera for High-Throughput Evaluation of Active Pharmaceutical Ingredients: A Case Study of Melting Point Determination

Kohsaku Kawakami[1,2,3]

Received 27 April 2010; accepted 23 July 2010; published online 31 July 2010

Abstract. Various techniques for physical characterization of active pharmaceutical ingredients, including X-ray powder diffraction, birefringence observation, Raman spectroscopy, and high-performance liquid chromatography, can be conducted using 96-well plates. The only exception among the important characterization items is the thermal analysis, which can be a limiting step in many cases, notably when screening the crystal/salt form. In this study, infrared thermal camera technology was applied for thermal characterization of pharmaceutical compounds. The melting temperature of model compounds was determined typically within 5 min, and the obtained melting temperature values agreed well with those from differential scanning calorimetry measurements. Since many compounds can be investigated simultaneously in this infrared technology, it should be promising for high-throughput thermal analysis in the pharmaceutical developmental process.

KEY WORDS: high-throughput analysis; infrared camera; thermal analysis.

INTRODUCTION

In the developmental study in pharmaceutical industry, once a candidate compound is identified for further development, its physicochemical characteristics must be extensively evaluated for designing clinical formulation. Notably, polymorphism of active pharmaceutical ingredients requires extensive investigation (1–4) because selection of an inadequate crystal form may cause serious, even fatal, problems during or after the launch of the products (5). However, since recent developmental timeframes in the pharmaceutical industry are much shorter than those of decades ago, various characterizations are carried out using high-throughput technologies (6–9). Various techniques for physical characterization, including X-ray powder diffraction (XRPD), birefringence observation, Raman spectroscopy, and high-performance liquid chromatography, are frequently conducted using 96-well plates. The only exception among the important characterization items is the thermal analysis, which can be a limiting step in many cases, notably when screening the crystal/salt form.

Infrared thermal camera technology provides with two-dimensional temperature images by detecting temperature-specific infrared energy from the samples. The field of this technology is rapidly expanding (10–14). For example, since recent instruments are very easy to handle, it is expected as a new methodology for diagnosis (13,14). Its application for thermal characterization of pharmaceutical compounds is introduced below.

MATERIALS AND METHODS

Materials

Indomethacin, carbamazepine, and griseofulvin were obtained from Wako Pure Chemicals (Osaka, Japan). Mannitol and sulfamerazine were supplied from Nacalai Tesque (Kyoto, Japan) and MP Biomedicals (Solon, OH, USA), respectively. Sulfamerazine from the supplier was in form I, which is the stable form at higher temperatures. It was transformed into form II, which is the stable form at lower temperatures, using a procedure described previously (15). All other reagents were of the highest grade available and used as received.

X-Ray Powder Diffraction

XRPD patterns were acquired on Rigaku RINT Ultima X-ray Diffraction System (Rigaku Denki, Tokyo, Japan) using CuKα radiation. The voltage and the current were 40 kV and 40 mA, respectively. The data were collected between 5° and 40° (two theta values) at intervals of 0.02° with a scan speed of 2°/min.

[1] National Institute for Materials Science, Biomaterials Center, Ibaraki, 305-0044, Japan.
[2] International Center for Materials Nanoarchitectonics, 1-1 Namiki, Tsukuba, Ibaraki, 305-0044, Japan.
[3] To whom correspondence should be addressed. (e-mail: kawakami.kohsaku@nims.go.jp)

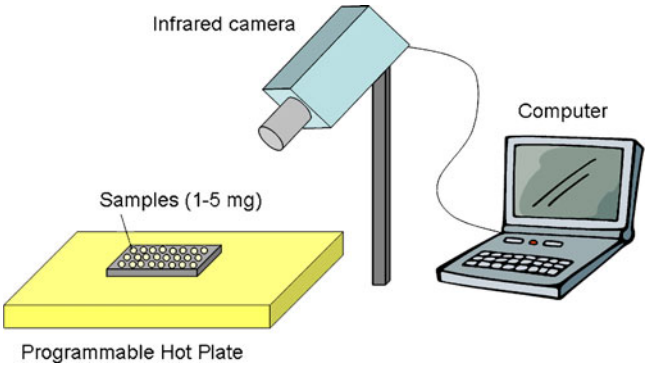

Fig. 1. Schematic representation of the experimental setup

Differential Scanning Calorimetry

Differential scanning calorimetry (DSC) measurements were performed on a Mettler Toledo DSC823^e (Mettler Toledo, Greifensee, Switzerland), which is periodically calibrated with Indium. About 4 (±1) mg of the sample was investigated using a sealed aluminum pan. Dry nitrogen (purity higher than 99.99%) was used as the inert gas at a flow rate of 30 ml/min. The heating rate was 15°C/min for all the samples.

Infrared Thermal Camera System

Figure 1 shows a schematic representation of the experimental setup. Each sample was heated on a programmable hot plate (Hakko Hot Plate DEMO, Hakko Electric Machine Works, Chikuma, Japan) with precise temperature control. All the samples were heated from 100°C with the linear ramp rate at 15°C/min in this study. The surface temperatures of the samples and the sample plate were investigated using an infrared thermal camera (FSV-7000E, Apiste, Osaka, Japan), with a temperature accuracy of ±2% and effective pixels of 320×240. Detection wavelength was 8–14 μm. Emissivity was the only parameter for the temperature calibration of the infrared camera. It could be regarded as almost constant in this study as described later. A black stainless steel plate was used as the sample plate, since reflection of the light disturbed the observation. The distance between the camera and the sample plate was approximately 30 cm. All the measurements were performed in an ambient atmosphere, and the data obtained was analyzed using the software from the manufacturer.

RESULTS AND DISCUSSION

Figures 2 and 3 show XRPD patterns and DSC curves of the model compounds used in this study. Indomethacin and mannitol were in the form γ and the form β, respectively, both of which have been recognized as the most stable forms thermodynamically (16–18). Carbamazepine was in the metastable state, the form III, which transformed into the most stable form I after the melting at 175°C (19,20). Only one form has been known for griseofulvin. Sulfamerazine has enantiotropically related two crystal forms (4,15,21). The intact form from the supplier (form I) was converted into the stable form at lower temperature (form II) in this study. In the heating process, it was transformed into the form I at 185°C, followed by the melting at 236°C.

Examples of thermal images are presented in Fig. 4, where five model compounds were heated at 15°C/min. Due to the low thermal conductivity of the solid state, the temperatures of the sample surfaces were initially lower than that of the sample plate, and thus the drug powders could be observed as distinct spots. Over time, the spots disappeared at the respective melting temperatures because the thermal conductivity of the resultant liquid was much higher to provide with the sample-surface temperature equal to that of the surrounding plate. Accordingly, the melting temperature of each sample could be obtained from the thermal image data.

Emissivity of the samples is required to calculate absolute temperatures. That of the pharmaceutical powders was about 0.86 in most cases, and this value can be used at least for the screening purposes. Figure 5 shows the surface temperatures of indomethacin and carbamazepine determined using the infrared camera. In the case of indomethacin, before melting, the surface temperature of the samples increased at a heating rate identical to that of the sample plate, although the temperature itself was lower than that of the surrounding plate. This was followed by a drastic increase in the sample temperature, caused by an increase in the thermal conductivity due to the phase transition from the solid to the liquid state. The endpoint of this rapid increase

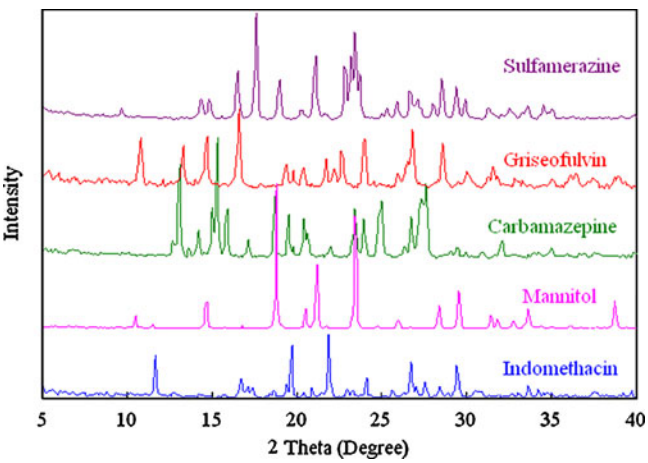

Fig. 2. XRPD patterns of the model compounds

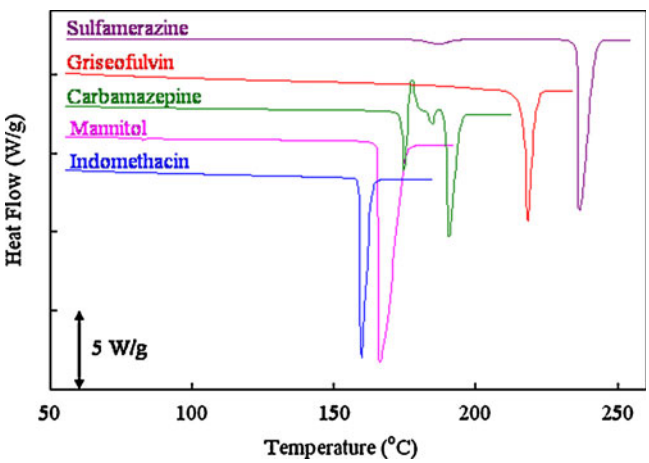

Fig. 3. DSC curves of the model compounds. Heating rate was 15°C/min

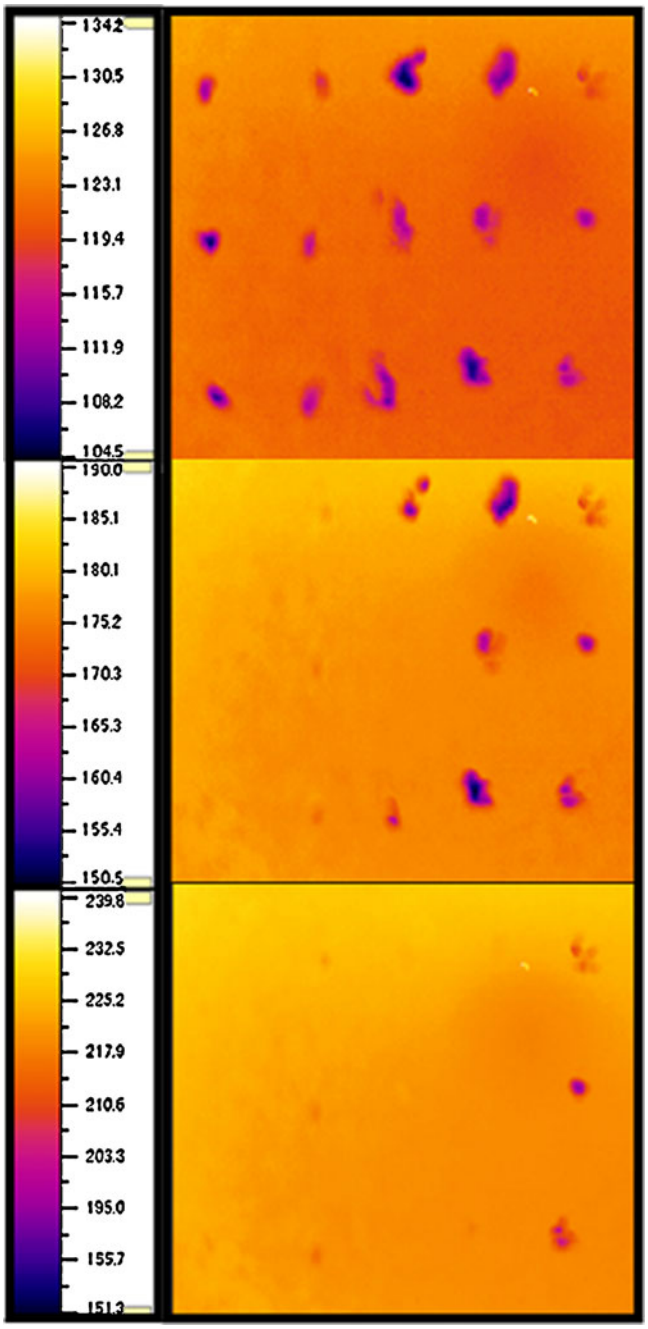

Fig. 4. Thermal images acquired over time using the infrared camera (*top* to *bottom*). The *left columns* indicate the temperature. The model drugs used were indomethacin (melting temperature, 160°C), mannitol (166°C), carbamazepine (174°C, 190°C), griseofulvin (219°C), and sulfamerazine (236°C; *left* to *right*, n=3)

could be regarded as the melting temperature of the sample. A difference in the sample amount as well as a difference in the degree of the thermal contact between the sample and the plate may have led to differences in the sample-surface temperature where the melting started, as shown in Fig. 5, but the melting temperatures were quite reproducible. The temperatures of the sample surface after melting were consistent with that of the sample plate. Moreover, the melting temperature determined using this method agreed with the value determined from the DSC measurement. This was also the case for mannitol and griseofulvin, of which the thermal behavior is very simple, that is, the melting is the only thermal event observed during the heating. Observation of carbamazepine was relatively difficult, which exhibited melt crystallization before reaching the melting point of the stable form. The first melting of the metastable carbamazepine at 174°C was detectable in the same manner as in the case of indomethacin, although clearness of the inflection was reduced due to subsequent recrystallization into the stable form. The second melting was also indicated at 190°C; however, it could not be detected sometimes. Since the problem was not the reproducibility in the temperature but the clearness of the inflection, it could be overcome by investigating multiple samples. Regarding sulfamerazine, although there is a polymorphic transition at about 185°C, it was not detectable.

An increase in the heating rate decreased the resolution as in the case of normal DSC measurement. On the other hand, there were no disadvantages in decreasing the heating rate except for the increase in the required measurement time. Although the decrease in sensitivity is a serious problem in the normal DSC measurement at slow heating rates, the proposed infrared technique is free from such a problem. Thus, the increase in the resolution can be achieved by decreasing the heating rate without decreasing the sensitivity. As for the sample

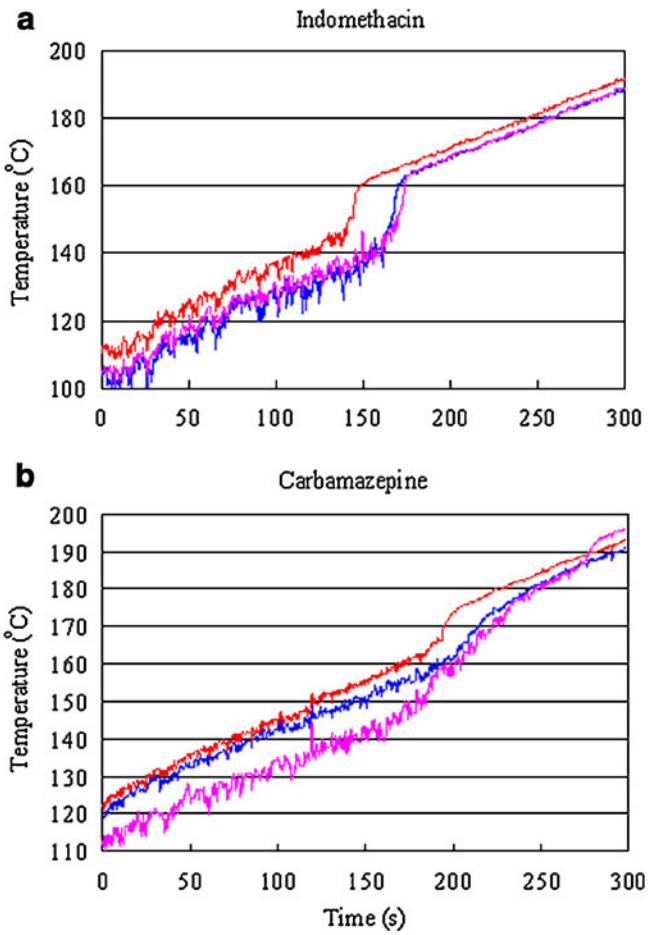

Fig. 5. Surface temperatures of **a** indomethacin and **b** carbamazepine acquired on the infrared camera. Three runs are differentiated by the *colored lines*

amount, although 1 mg was usable for the observation, it decreased the temperature difference between the sample surface and the surrounding plate, which reduced clearness of the inflection in the temperature–time plot. Thus, at least use of 3 mg of the sample is recommended for the observation.

The melting temperature has been the only thermal event clearly observed in this study. However, an increase in the sensitivity should enable observation of any thermal events, including polymorphic transition, desolvation, and crystallization, because any events accompany thermal responses. Needless to say, factors that can cause problems in the normal DSC measurements, such as large amount of impurities and contamination of the different crystal forms, should be problems in this analysis as well. The factor relating to a decrease in the sensitivity of the proposed method is, for example, noise from the air, which should be overcome by reducing the atmospheric pressure. It should be noted that the measurements shown in Fig. 5 were finished within 5 min. It may be possible to observe 96 samples in a few minutes by heating a 96-well plate at a higher heating rate. This application of infrared technology is a promising methodology for high-throughput thermal analysis in the pharmaceutical developmental process.

CONCLUSIONS

Infrared thermal camera technology was employed for thermal characterization of pharmaceutical compounds. The melting temperature of model compounds was typically determined within 5 min, and it may become shorter with higher heating rate. The melting temperature values, which were determined from the temperature–time plots, agreed well with those from DSC measurements. Since many compounds can be investigated simultaneously in this infrared technology, it should be promising for high-throughput thermal analysis in the pharmaceutical developmental process.

ACKNOWLEDGMENT

The author would like to thank Apiste (Osaka, Japan) for the use of the infrared camera. This work was in part supported by World Premier International Research Center (WPI) Initiative on Materials Nanoarchitectonics, MEXT, Japan.

REFERENCES

1. Hilfiker R, Blatter F, von Raumer M. Relevance of solid-state properties for pharmaceutical products. In: Hilfiker R, editor. Polymorphism. Weinhelm: Wiley-VCH; 2006. p. 1–19.
2. Pudipeddi M, Serajuddin ATM. Trends in solubility of polymorphs. J Pharm Sci. 2005;94:929–39.
3. Miller JM, Collman BM, Greene LR, Grant DJW, Blackburn AC. Identifying the stable polymorph early in the drug discovery-development process. Pharm Dev Technol. 2005;10:291–7.
4. Kawakami K. Reversibility of enantiotropically related polymorphic transformations from a practical viewpoint: thermal analysis of kinetically reversible/irreversible polymorphic transformations. J Pharm Sci. 2007;96:982–9.
5. Bauer J, Spanton S, Henry R, Quick J, Dziki W, Porter W, *et al*. Ritonavir: an extraordinary example of conformational polymorphism. Pharm Res. 2001;18:859–66.
6. Newman AW, Childs SL, Cowans BA. Salt and cocrystal form selection. In Preclinical development handbook. Hoboken: Wiley; 2008. Chapter 14.
7. Hilfiker R, De Paul SM, Szelagiewicz M. Approaches to polymorphism screening. In: Hilfiker R, editor. Polymorphism. Weinhelm: Wiley-VCH; 2006. p. 287–308.
8. Henck JO, Byrn S. Designing a molecular delivery system within a preclinical timeframe. Drug Discovery Today. 2007;12:189–99.
9. Kawakami K. Current status of amorphous formulation and other special dosage forms as formulations for early clinical phases. J Pharm Sci. 2009;98:2875–85.
10. Telenkov SA, Vergas G, Nelson JS, Milner TE. Coherent thermal wave imaging of subsurface chromophores in biological materials. Phys Med Biol. 2002;47:657–71.
11. Hashimoto T, Morikawa J. Two-dimensional microscale thermal analysis of freezing of onionskin cells by high-speed infrared focal plane arrays. J Appl Phys. 2003;42:L706–8.
12. Meola C, Carlomagno GM. Application of infrared thermography to adhesion science. J Adhesion Sci Technol. 2006;20:589–632.
13. Ng EYK. A review of thermography as promising non-invasive detection modality for breast cancer. Int J Therm Sci. 2009;48:849–59.
14. Tan JH, Ng EYK, Rajendra Acharya U. Infrared thermography on ocular surface temperature: a review. Infrared Phys Technol. 2009;52:97–108.
15. Kawakami K, Ida Y. Application of modulated-temperature DSC to the analysis of enantiotropically-related polymorphic transitions. Thermochim Acta. 2005;427:93–9.
16. Crowley KJ, Zografi G. Cryogenic grinding of indomethacin polymorphs and solvates: assessment of amorphous phase formulation and amorphous phase physical stability. J Pharm Sci. 2002;91:492–507.
17. Kett VL, Fitzpatrick S, Cooper B, Craig DQM. An investigation into the subambient behavior of aqueous mannitol solutions using differential scanning calorimetry, cold stage microscopy, and x-ray diffractometry. J Pharm Sci. 2003;92:1919–29.
18. Xie Y, Cao W, Krishnan S, Lin H, Cauchon N. Characterization of mannitol polymorphic forms in lyophilized protein formulations using a multivariate curve resolution (MCR)-based Raman spectroscopic method. Pharm Res. 2008;25:2292–301.
19. Kobayashi Y, Ito S, Itai S, Yamamoto K. Physicochemical properties and bioavailability of carbamazepine polymorphs and dehydrate. Int J Pharm. 2000;193:137–46.
20. Grzesiak AL, Lang M, Kim K, Matzger AJ. Comparison of the four anhydrous polymorphs of carbamazepine and the crystal structure of form I. J Pharm Sci. 2003;92:2260–71.
21. Zhang GGZ, Gu C, Zell MT, Burkhardt RT, Munson EJ, Grant DJW. Crystallization and transitions of sulfamerazine polymorphs. J Pharm Sci. 2002;91:1089–100.