Journal of Pharmaceutical and Biomedical Analysis 84 (2013) 177–183

Contents lists available at SciVerse ScienceDirect

Journal of Pharmaceutical and Biomedical Analysis

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

Polymorph screening of an active material

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ARTICLE INFO

Article history:
Received 19 March 2013
Received in revised form 31 May 2013
Accepted 3 June 2013
Available online 14 June 2013

Keywords:
Polymorph
Drug candidate
Relative stability
Dissolution test

ABSTRACT

Polymorph screening is currently one of the most important tasks for innovators and for generic companies from both pharmaceutical and intellectual property rights aspects. The different polymorphs have different physicochemical properties, such as the crystal polymorph-dependent solubility which influences the bioavailability.

A former drug candidate obtained from Sanofi Pharmaceutical Company (Hungary) was investigated to explore its polymorphism, to distinguish the morphologies generated by analytical examinations and to investigate their relative stabilities. An Avantium Crystal 16 automatic laboratory reactor system was used for the polymorph studies and the studies of their dissolution. Eight polymorphs were obtained by crystallization and transformation methods then characterized by XRPD, DSC, and Raman spectroscopy, scanning electron microscopy, and light microscopy. All the morphologies could be stored in solid without any form transformation for a long time (2 years investigated). According to the first relative stability results, Form I, III, IVa, V, VI, VII are unambiguously metastable forms. Form II and IVb have similar thermodynamic stabilities, that were higher than those of the other polymorphs.

A special dissolution medium was developed in which the eight polymorphs showed clear differences in the rate of dissolution.

1. Introduction

Polymorphism, the ability of an element or compound to crystallize in more than one distinct crystal species [1], is a major problem that has been of considerable importance in the pharmaceutical industry in the development of new drug candidates [2,3]. The different polymorphs exhibit different physical and physicochemical properties, such as habit, colour, density, melting point, solubility, dissolution rate, etc. [4]. These differences may affect the pharmaceutical processing, the stability of the drug product, the bioavailability and the toxicity, and thereby the therapeutic efficacy of the drug substance [5]. Furthermore, each crystal phase can be protected by patents [6], which is very important for the innovator. During the polymorphism screening process, it is necessary to investigate the relative stability, because mostly thermodynamically stable forms are used in the drug products [7].

The most common polymorph screening methods are crystallization from melt, vapour, and solutions through cooling or evaporating the solvent, addition of an antisolvent to the solution [8], or slurrying the solid active pharmaceutical ingredient for an extended period of time at different temperatures [9].

The outcome of a crystallization can be affected by factors such as the solvent (viscosity, polarity), the rate of creating supersaturation (cooling, adding or evaporation rate), or initial solution concentration [10]. Other crystallization conditions, e.g. the rate of stirring, different mechanical processes (e.g. ultrasound effect) or different additives, may also influence the final crystal polymorph obtained.

The most common methods for the characterization of the various polymorphs are powder X-ray diffractomerty (XRPD), single-crystal X-ray diffractometry (SC-XRD), differential scanning calorimetry (DSC), optical and electron microscopy, infrared (IR), near-infrared (NIR), Raman, and more recently solid-state nuclear magnetic resonance spectroscopy (ssNMR) [11–16].

In case of polymorphism the relative stability and the transformation conditions of the different polymorphs must be investigated [17]. Polymorphic transformations between crystalline modifications of an active pharmaceutical ingredient in, e.g. crystallization, heating/cooling studies and milling have been already characterized by various solid-state analytical methods [18]. An Avantium Crystal 16 automatic laboratory reactor system was used for such investigations. This medium-throughput polymorph and salt screening technology performs 16 parallel crystallization experiments, and provides an estimate of solubility by using turbidity measurements [19]. With this reactor system, the solubility and supersolubility curves can also be determined [20].

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http://dx.doi.org/10.1016/j.jpba.2013.06.002

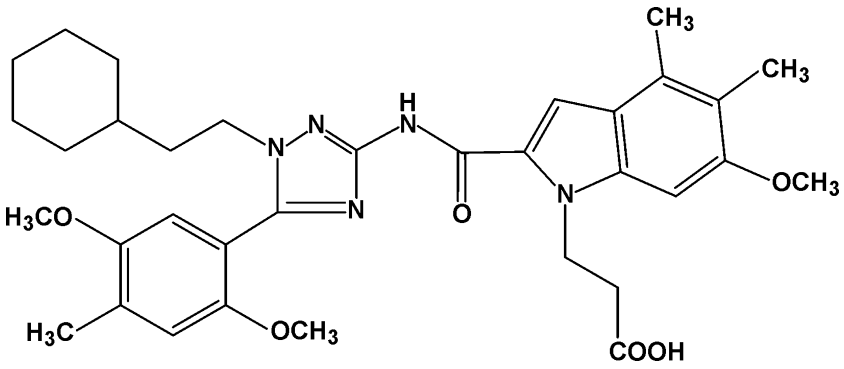

Fig. 1. Chemical structure of the former drug candidate.

The model compound of this study was a former drug candidate obtained from Sanofi Pharmaceutical Company. Here, a description of the polymorphism of a drug candidate is presented without using additives during the generating process. The polymorphs were generated by crystallization methods (cooling, evaporation) and heating transformation, furthermore the modifications obtained were distinguished by means of various analytical examinations. For the dissolution study, a special dissolution medium was developed in which the different polymorphs could be distinguished.

2. Experimental

2.1. Materials

The model drug used was 3-[2-({[1-(2-cyclohexylethyl)-5-(2,5-dimethoxy-4-methylphenyl)-1H-1,2,4-triazol-3-yl]amino}carbonyl)-6-methoxy-4,5-dimethyl-1H-indol-1-yl}propanoic acid (Fig. 1). The crystallization of the various polymorphic forms was achieved with different organic solvents of analytical grade: acetonitrile, methanol, 96% ethanol, abs. ethanol, isopropanol, acetone, 2-butanone, toluene (Merck, Budapest, Hungary), dichloromethane, ethyl acetate, butyl acetate, chloroform, 1,4-dioxane (Reanal, Budapest, Hungary), and tetrahydrofuran (Aldrich, Budapest, Hungary).

2.1.1. Preparation of polymorphs by crystallization

Different techniques were used for the preparation of the polymorphs (Table 1):

Crystallization by shock cooling: The raw material was dissolved at the boiling point of the solvent to give a saturated solution which was then diluted with a small amount of the solvent. The hot solution was filtered into a vial immersed into crashed ice.

Crystallization by slow cooling: The same process of dissolution was used, but the filtered, hot solution was cooled down slowly (−3 °C/h).

Crystallization by slow evaporation: The products were the dried residues of the clear filtrates of the samples prepared by shock cooling.

2.1.2. Preparation of polymorphs by heating transformation

Through heating processes at various temperatures for different periods of time, the polymorphs generated by crystallization could be transformed to other forms.

2.2. Investigation methods

2.2.1. Identification of crystalline forms of samples

2.2.1.1. Powder X-ray diffractometry. XRPD spectra were recorded with a BRUKER D8 Advance diffractometer (Bruker AXS GmbH, Karlsruhe, Germany) system with Cu $K\alpha_1$ radiation ($\lambda = 1.5406$ Å) over the interval 2.5–40°/2θ. The measurement conditions were as follows: target, Cu; filter, Ni; voltage, 40 kV; current, 40 mA; time constant, 0.1 s; angular step 0.016°. Detecor: NaI (Tl) scintillation detector.

2.2.1.2. Differential scanning calorimetry. DSC curves were obtained by a Mettler Toledo DSC27HP apparatus (Mettler-Toledo AG, Greifensee, Switzerland) under the following conditions: sample weight: about 2–3 mg; sample holder: aluminum crucible (40 μL) with lid; nitrogen flow rate: 100–150 mL/min; heating rate: 10 °C/min from 25 °C up to 250 °C. In every case, samples were held at 25 °C for 10 min before recording.

2.2.1.3. Raman spectroscopy. Raman spectra were recorded at room temperature by using a Bruker SENTERRA Dispersive Raman Microscope (Bruker Optik GmbH, Ettlingen, Germany) equipped with Nd-YAG (532 nm) and diode (785 nm) excitation lasers and a cooled CCD detector. The system was fitted with a motorized XYZ sample stage. The equipment was controlled by OPUS 6.5 software (Bruker Optik GmbH, Ettlingen, Germany). Samples were analyzed on glass slides.

Following a parameter optimization process, the samples were measured by Nd-YAG (532 nm) laser at a laser power of 10 mW. An integration time of 5 s, and 3 scans were used for each measurement, and spectra were collected over the Raman shift range of 3500–60 cm^{-1} at a resolution of 3–5 cm^{-1}. The microscope magnification was 200× and a certain crystal was selected for analysis with

Table 1
The techniques and conditions used for preparation of the polymorphs.

Form	Method	Solvent	Conditions
I	Crystallization	96%, abs. ethanol	Shock cooling
II	Heating	–	160 °C/6 h (from Form I)
III	Crystallization	Isopropanol, 2-butanone, butylacetate, ethylacetate, toluene	Shock cooling
IVa	Crystallization	Methanol	Shock cooling
IVb	Heating	–	205 °C/6 h (from Form IVa)
V	Crystallization	Chloroform	Shock cooling, slow cooling, slow evaporation
VI	Crystallization	Acetone, methanol, dichloromethane, 1,4-dioxane	Shock cooling, slow cooling, slow evaporation
VII	Crystallization	Acetonitrile	Shock cooling