

# The Development of Quasi-isothermal Calorimetry for the Measurement of Drug–Polymer Miscibility and Crystallization Kinetics: Olanzapine-Loaded PLGA Microparticles

Sean Askin,<sup>†</sup> Min Zhao,<sup>‡</sup> Andrea D. Gonçalves,<sup>§</sup> Simon Gaisford,<sup>†</sup> and Duncan Q. M. Craig<sup>\*,†</sup>

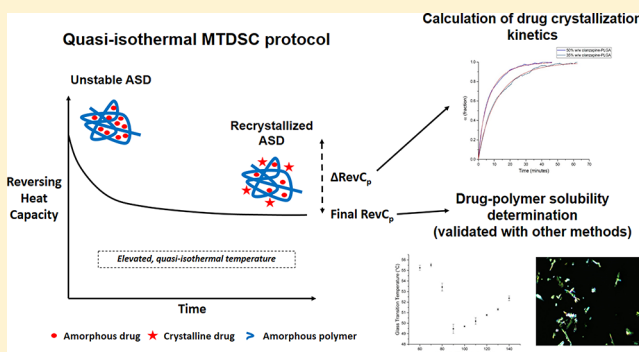
<sup>†</sup>UCL School of Pharmacy, 29-39 Brunswick Square, London WC1N 1AX, United Kingdom

<sup>‡</sup>School of Pharmacy, Queen's University Belfast, 97 Lisburn Road, Belfast BT9 7BL, United Kingdom

<sup>§</sup>DPDD Drug Delivery, GlaxoSmithKline R&D, Gunnels Wood Road, Stevenage SG1 2NY, United Kingdom

**ABSTRACT:** The assessment of drug–polymer equilibrium solubility is of critical importance for predicting suitable loading and physical stability of solid dispersion formulations. However, quantitative measurement of this parameter is nontrivial due to the difficulties associated with ascertaining equilibrium values in systems that are prone to supersaturation and are simultaneously highly viscous, thereby slowing the equilibration process considerably; no standard methodology has yet been agreed for such measurements. In this study, we propose a new approach involving quasi-isothermal modulated temperature DSC (QiMTDSC), whereby unsaturated and supersaturated samples are held at defined temperatures and subject to a sinusoidal heating signal at a zero underpinning heating rate, thereby allowing the heat capacity of the sample to be measured as a function of time and temperature. We are not only able to ascertain whether equilibrium has been reached by monitoring the time-dependent heat capacity signal, but we can also measure solubility as a function of temperature via the absolute heat capacity values of the components. We are also able to measure the kinetics of recrystallization from the supersaturated systems. Dispersions of olanzapine in PLGA at concentrations up to 50% w/w, prepared by spray drying, were prepared and characterized using conventional and QiMTDSC as well as hot stage microscopy. The new QiMTDSC protocol was successfully able to determine olanzapine solubility in PLGA at 90 °C to be  $23.1 \pm 6.1\%$  w/w, which was comparable to the values calculated using other established methods at this temperature, while a temperature/solubility profile was obtained using the method at a range of temperatures. Drug crystallization kinetics from the solid dispersions could also be modeled directly from the QiMTDSC data using the Avrami approach, thereby allowing the effect of drug loading on the rate of crystallization and the effective completion of crystallization to be investigated. Overall, an alternative protocol for measuring drug–polymer solubility has been developed and validated via comparison to established methods, the approach allowing solubility as a function of temperature, identification of equilibrium following demixing, and kinetic analysis of crystallization to be performed within one set of experiments.

**KEYWORDS:** drug–polymer solubility, amorphous solid dispersion, PLGA, microparticles, olanzapine, crystallization, crystallization kinetics, differential scanning calorimetry, quasi-isothermal modulated temperature differential scanning calorimetry, polarized light microscopy, physical stability, demixing



## INTRODUCTION

Amorphous solid dispersions of drug molecules in a polymer matrix are a well-established approach to achieve both controlled release or solubilization.<sup>1</sup> The amorphous form of most small molecule drugs is inherently unstable, and the physical stability of such systems is determined by the thermodynamic equilibrium solubility level of the drug in the polymer, which determines the maximum drug load that can be molecularly dispersed in the polymer matrix without risk of phase separation over time. Drug–polymer solubility is temperature-dependent and is influenced by both the enthalpy and entropy of mixing, although the relative contributions of

each are uncertain.<sup>2</sup> The capability to understand the physical stability of a formulation is essential as changes in the physical state of the drug, such as crystallization, can significantly affect the release performance of the solid dispersion, which makes determining the solubility of the drug in the polymer of critical importance.<sup>3</sup> Despite this, there is still no standard method for measuring drug–polymer solubility, because there are several

**Received:** April 5, 2018

**Revised:** June 19, 2018

**Accepted:** June 22, 2018

**Published:** June 22, 2018



difficulties associated with making solubility measurements in polymers, which are a result of the high viscosity of these systems. The main issues with the high viscosity of the polymer matrix are that the solubility equilibrium kinetics are very slow, and quantification of the drug concentration in the solid polymer matrix is challenging.

Modeling and experimental approaches have both been used to investigate the thermodynamic drug–polymer solubility equilibrium. Perturbed-chain statistical associating fluid theory (PC-SAFT) has been used to thermodynamically model the effect of polymer composition and molecular weight on solubility in drug–polymer systems and has been shown to provide accurate predictions of drug–polymer solubility through comparison to experimental results.<sup>4–6</sup> Experimental protocols for measuring drug–polymer solubility have traditionally used differential scanning calorimetry (DSC). DSC can use elevated temperatures, which allow the solubility equilibrium to be achieved more quickly, and can also measure key properties of the system, such as melting endotherms and glass transition temperatures, which can be used to determine solubility. A seminal DSC method reported in the literature involved heating cryo-milled physical mixtures of drug and polymer with the aim of dissolving the drug in the molten matrix by heating at slow scan rates and measuring at which temperature the dissolution process ends ( $T_{\text{end}}$ ).<sup>7</sup>  $T_{\text{end}}$  was therefore the temperature at which the solubility is equal to the weight fraction of the drug in the physical mixture for the system. The method was then further developed by Sun et al. to include annealing periods to allow more time for the equilibrium to be reached, and a fast scan rate was used to assess whether any crystalline material remained following annealing.<sup>8</sup>

More recently, Mahieu et al. developed a new DSC protocol based on demixing supersaturated drug–polymer solid dispersions.<sup>9</sup> This approach uses a heat–cool–heat protocol: In the first cycle, the solid dispersion is heated to promote crystallization of the thermodynamically unstable drug and is then annealed at a defined temperature to allow the solubility equilibrium to be achieved. In the second cycle, the sample is rapidly cooled below the glass transition temperature ( $T_g$ ). Finally, in the third cycle, the sample is heated again, and the  $T_g$  of the molecularly dispersed drug–polymer phase is measured. The composition of the drug–polymer phase can then be determined from the  $T_g$  using theoretical relationships such as the Gordon–Taylor equation.<sup>10</sup> A key advantage of this approach is that the demixing process (crystallization) is often faster than the mixing process (dissolution) on heating of these highly viscous systems, which makes experimental times significantly shorter.<sup>9</sup> This method has now been applied to several drug–polymer systems and has been validated against other methods of measuring solubility.<sup>11–15</sup>

In this study, a new quasi-isothermal modulated temperature DSC (QiMTDSC) protocol has been developed as a novel means to simultaneously measure both drug–polymer solubility and drug crystallization kinetics within solid dispersions based upon the same demixing principle. QiMTDSC is a variant of traditional MTDSC, which uses an underlying heating rate of zero. The sample temperature is modulated around a central value for extended periods, which allows the reversing heat capacity ( $\text{Rev}C_p$ ) of a sample to be measured as a function of time. Heat capacity is a fundamental measure of molecular mobility. At temperatures below the  $T_g$ , the glassy and crystalline forms of a material have a very similar

heat capacity; however, above the  $T_g$ , the amorphous form of a material has a significantly higher heat capacity than the crystalline counterpart due to changes in the configurational degrees of freedom.<sup>16</sup> The newly developed QiMTDSC method therefore proposes to exploit the heat capacity difference between the two physical forms of the drug above the  $T_g$  to both determine the fraction of drug that crystallizes at a particular temperature (therefore allowing solubility calculations) and measure the kinetics of the drug crystallization that occurs in the solid dispersion system.

Olanzapine and poly(lactic-co-glycolic acid) (PLGA) form the system of interest in this study. Olanzapine is an atypical antipsychotic medication licensed for maintenance treatment of schizophrenia.<sup>17</sup> PLGA is a synthetic biodegradable polyester that is widely used for long-acting formulations such as microparticles because of its proven safety in humans and long in vivo degradation times, which allow controlled release of drug over time periods of days to years.<sup>18</sup> Several studies have previously formulated olanzapine with PLGA to produce micro/nanoparticles, because long-acting injectable formulations of olanzapine are desirable due to issues with compliance to oral therapy of antipsychotic medications in patients with schizophrenia.<sup>19–22</sup> No attempts have yet been made to measure the drug–polymer solubility of this formulation, which may have a significant effect on drug loading and release performance.

The aims of this study were three-fold: (1) to measure olanzapine–PLGA solubility using the established DSC demixing method, (2) to develop a QiMTDSC protocol as a novel method to both measure drug–polymer solubility and drug crystallization kinetics within solid dispersion systems, and (3) to compare the drug–polymer solubility values calculated using the calorimetry methods to a simple microscopy method at a single temperature. This study therefore provides a comparison of three demixing methods used to assess drug–polymer solubility and also describes the development of a new QiMTDSC protocol for investigating the thermodynamic and kinetic stability of amorphous solid dispersion formulations.

## ■ EXPERIMENTAL SECTION

**Materials.** Crystalline olanzapine ( $\text{C}_{17}\text{H}_{21}\text{ON}_4\text{S}$ ;  $M_w = 312.43$  g/mol) was purchased from Mjoy Ltd., India. Amorphous PLGA (lactide/glycolide ratio = 50:50;  $M_w = 17\,000$  g/mol; acid terminated) was obtained from Corbion, Netherlands. Dichloromethane (DCM) and acetonitrile were of analytical grade and were purchased from VWR, United Kingdom.

**Methods. Preparation of Amorphous Microparticulate Solid Dispersion Formulations.** Olanzapine–PLGA microparticles (polymer dispersions) were produced using a Mini B-290 Spray Dryer (Buchi, Switzerland) connected to a B-295 Inert Loop (Buchi, Switzerland), achieving a closed system. The materials were codissolved in DCM at the desired ratio at a total solids concentration of 10% w/v. Spray drying was performed under an inert atmosphere (<6%  $\text{O}_2$ ), which was achieved by use of nitrogen spray gas. Atomization of the feed solution was achieved using an ultrasonic nozzle and controller (Buchi, Switzerland) with an operating power of 1.0 W and a pump rate of 1.25 mL/min. The temperature of the drying gas was 60 °C, resulting in an outlet gas temperature of 40–45 °C. The product obtained was a fine yellow powder, and a secondary drying step was performed in an oven (40 °C, 24 h)

to remove residual solvent. A UV assay ( $\lambda = 276$  nm) was used to quantify the drug content of the particles, which were first fully dissolved in acetonitrile. The measured drug content was in excellent agreement with the theoretical composition at all drug loads used in this study.

**Thermal Analysis.** The standard DSC experiments were performed using a DSC Q1000 (TA Instruments, USA), and the QiMTDSC experiments were performed using a Discovery DSC (TA Instruments, USA). All DSC experiments were performed under a nitrogen purge gas at a flow rate of 50 mL/min. Temperature and cell constant calibrations were performed on each instrument using indium as a standard. An additional reversing heat capacity ( $\text{RevC}_p$ ) calibration was performed on the Discovery DSC using synthetic sapphire as a standard. Standard aluminum pans and lids (TA Instruments, USA) were used for all experiments. A pinhole was punched in the lids of the pans used in the standard DSC experiments. A sample mass of  $5 \pm 0.5$  mg was used for all experiments. Additional care was taken when preparing samples for QiMTDSC to ensure that a uniform mass was present and the bottom of the DSC pan was completely flat. The  $\text{RevC}_p$  was also verified several times throughout the experiment.

**Optical Microscopy.** Polarized light microscopy was performed using a Leica DM 2700 M microscope (25 $\times$  magnification) with a polarized light filter and a Mettler Toledo HS82 Hot Stage controlled by an FP90 Central Processor.

**Methods for Measuring Drug–Polymer Solubility. DSC Demixing ( $T_g$ ) Protocol.** The protocol used in this study was similar to that described in the original publication<sup>9</sup> and is based on measuring drug–polymer solubility using the  $T_g$  after reaching the thermodynamic solubility equilibrium from a supersaturated state. Briefly, a 50% w/w olanzapine–PLGA solid dispersion was heated at a rate of 10 °C/min from 0 °C to the target temperature where it was annealed for 90 min. The samples were then rapidly cooled to  $-10$  °C and then reheated to 200 °C at a rate of 10 °C/min. The  $T_g$  of the amorphous phase following crystallization was measured in the second heating cycle. Experiments were performed in triplicate at each annealing temperature and an average  $T_g$  was calculated.

The  $T_g$  was used to calculate the solubility following crystallization using a standard curve as the system was found to significantly deviate from the theoretical Gordon–Taylor relationship. To prepare the standard curve, a series of olanzapine–PLGA solid dispersions with increasing drug content were produced using the spray drying method described above, and the  $T_g$  was measured using a standard heat–cool–heat method with a heating rate of 10 °C/min. The samples were heated to 70 °C in the first cycle and held at this temperature isothermally for a period of 30 min to thoroughly dry the solid dispersions before measuring the  $T_g$  in the second heating cycle.

**QiMTDSC Demixing ( $\text{RevC}_p$ ) Protocol.** A new QiMTDSC protocol was developed in this study that provides an alternative method for measuring drug–polymer solubility. This method also used the demixing principle to achieve the thermodynamic solubility equilibrium; however, the  $\text{RevC}_p$  of the system is used to calculate the solubility rather than the  $T_g$ .

The  $\text{RevC}_p$  of crystalline olanzapine, amorphous olanzapine, and amorphous PLGA were measured at 90 °C. Amorphous olanzapine was generated by first melting the crystalline drug in situ in the DSC pan. The materials were equilibrated to 90

°C and held isothermally for 15 min with a modulation amplitude of 1 °C and period of 100 s to measure the  $\text{RevC}_p$ . Six replicates of each material were analyzed, and an average value was used.

Olanzapine–PLGA microparticle solid dispersions (10, 35, and 50% w/w drug load) were then analyzed. The  $\text{RevC}_p$  of the samples were measured at 90 °C, as described above, using an isothermal period of 70 min to allow time for olanzapine crystallization. The  $\text{RevC}_p$  of the raw materials and the  $\text{RevC}_p$  of the solid dispersions following annealing could then be used to calculate the relative fractions of crystalline and amorphous drug, which could then be used to calculate the drug–polymer solubility.

Additionally, this method allowed the kinetic stability of supersaturated systems to be investigated. The olanzapine crystallization kinetics were calculated for each solid dispersion system by fitting the Avrami equation to the experimental data using the Levenberg–Marquardt iteration algorithm in Origin Pro 2016 software.

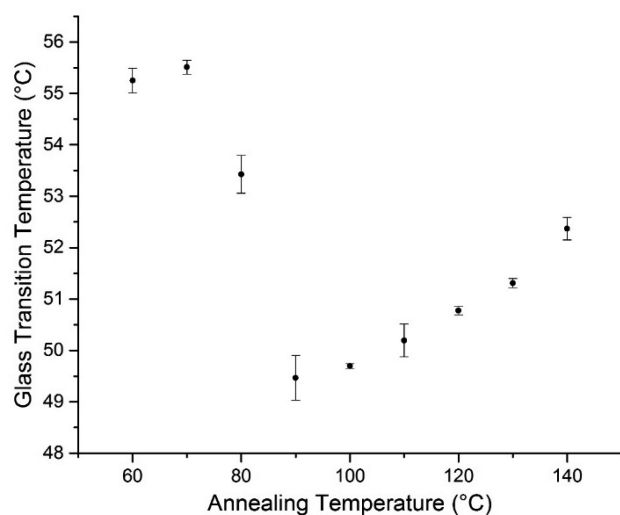
A 50% w/w olanzapine–PLGA microparticle solid dispersion was also analyzed at 70, 80, and 90 °C for 90 min using the same modulation parameters to assess whether crystallization was complete under these annealing conditions for comparison to the DSC ( $T_g$ ) protocol data.

**Semiquantitative Microscopy Method.** A semiquantitative microscopy method was used to verify the olanzapine–PLGA solubility values calculated using the calorimetry methods. A series of olanzapine–PLGA solid dispersions with increasing drug load were prepared by spray drying. Each solid dispersion (<1 mg) was then finely spread on a glass microscope slide and stored at 90 °C in an oven for 24 h to promote demixing (crystallization) and allow time for the solubility equilibrium to be achieved. The annealed solid dispersions were then removed from the oven and placed in a hot stage set at 90 °C, and polarized light microscopy was used to assess for the presence of olanzapine crystals, indicating whether the system was undersaturated or supersaturated at the temperature in question.

## ■ RESULTS

**Measuring Drug–Polymer Solubility: DSC Demixing ( $T_g$ ) Method.** In the first part of the study, highly loaded systems were annealed in order to promote demixing, with the measurement of postphase separation  $T_g$  used as a means of assessing the remaining drug concentration, which is considered to be the saturation drug level. The  $T_g$  values of amorphous olanzapine and PLGA were measured as 68.1 and 40.4 °C, respectively. A 50% w/w olanzapine–PLGA microparticle solid dispersion was then annealed at a range of temperatures, and the  $T_g$  was subsequently measured on reheating following cooling under controlled conditions. Figure 1 shows the average  $T_g$  measured versus annealing temperature for the temperature range of 60–140 °C. The  $T_g$  of the solid dispersion remains fairly constant at approximately 55.2 °C following annealing at 60 and 70 °C, suggesting that the system does not demix under these conditions. However, the  $T_g$  begins decreasing when annealed at 80 °C and decreases further to 49.5 °C when annealed at 90 °C, reflecting the loss of drug from the amorphous phase as it crystallizes, with an associated decrease in  $T_g$  as the PLGA content effectively increases in the amorphous phase. This reduction in  $T_g$  between 70 and 90 °C coincided with a reduction in enthalpy in the crystallization exotherm (Figure 2), which

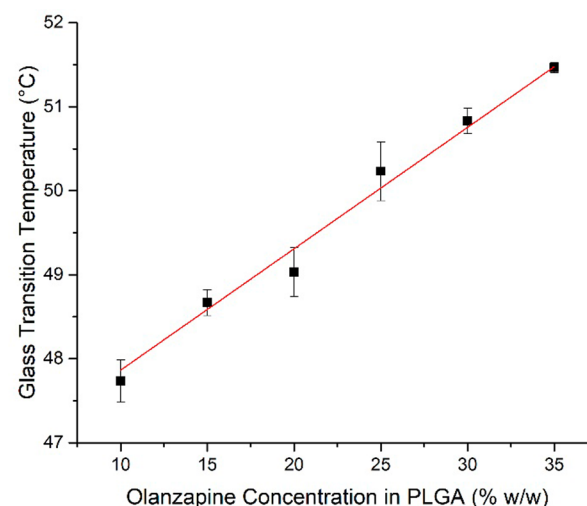




**Figure 1.** Effect of annealing temperature on the glass transition of olanzapine–PLGA 50% w/w microparticles

confirms that reduction in  $T_g$  is due to loss of amorphous olanzapine from the polymer matrix as it crystallizes during the annealing period. The lowest annealing temperature at which no crystallization was detected in the second heating cycle was 90 °C, indicating that for the system and annealing time used, 90 °C is the lowest temperature at which complete demixing occurs. Therefore, temperatures  $\geq 90$  °C can be used to measure solubility as the thermodynamic solubility equilibrium is achieved. Annealing in the temperature range between 100 and 140 °C resulted in a gradual increase in the  $T_g$  to 52.4 °C (Figure 1). An increase in  $T_g$  can be interpreted in terms of the solubility being temperature-dependent; hence, as the temperature increases, the excess crystalline drug dissolves. The drug–polymer solubility can therefore be calculated at a range of temperatures, and a solubility curve can be generated at elevated temperatures.

The solubility was calculated from the  $T_g$  of the solid dispersion using a standard curve of  $T_g$  versus olanzapine concentration in PLGA (Figure 3). The standard curve was

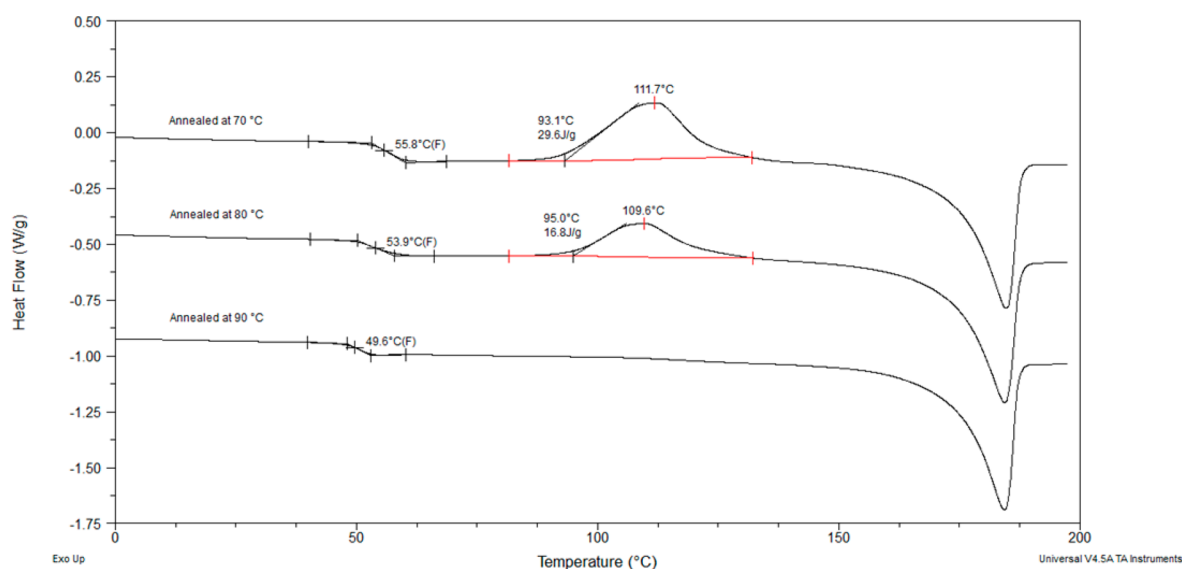


**Figure 3.** Standard curve of  $T_g$  of solid dispersion vs olanzapine concentration in PLGA used to calculate solubility from  $T_g$  due to deviation of the system from the Gordon–Taylor relationship.

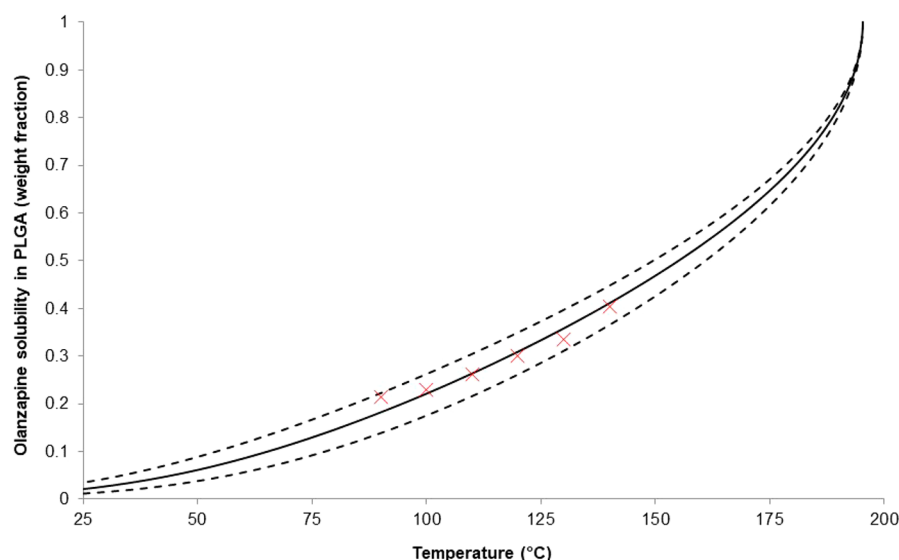
fitted with a linear trend line ( $R^2 = 0.987$ ) to allow calculation of olanzapine solubility in PLGA from the  $T_g$ ; this relationship was found to provide a better fit than the Gordon–Taylor equation in this case. The solubility values calculated using this method are displayed in Table 1 and have been used to

**Table 1.** Olanzapine–PLGA Solubility Calculated from DSC Demixing Method Using  $T_g$  Measurements

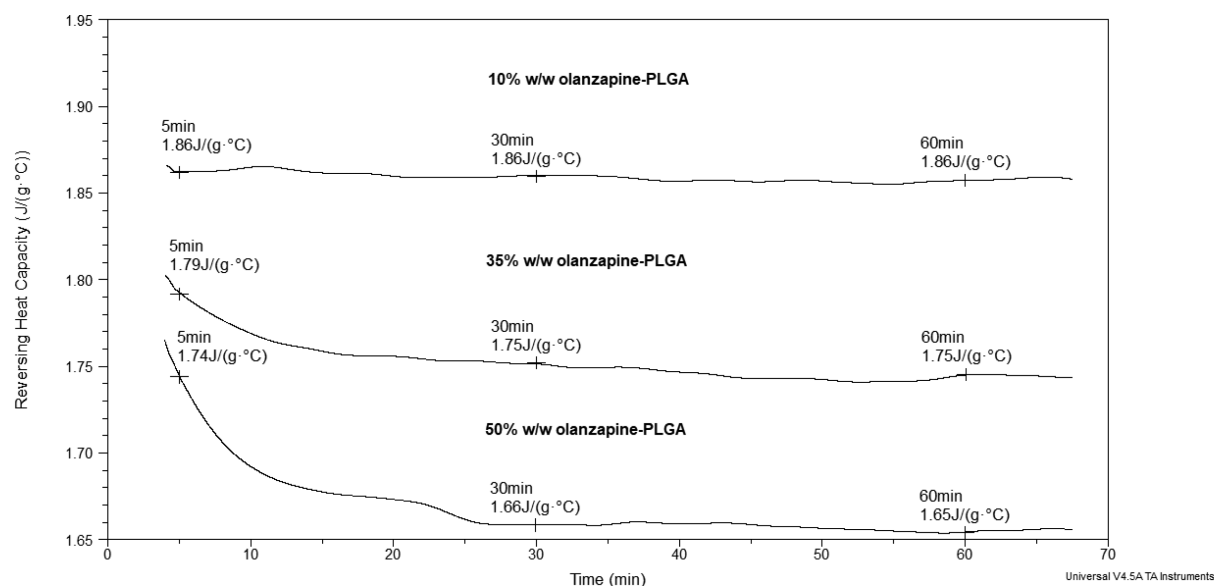
annealing temperature (°C)	$T_g \pm$ standard deviation (°C)	average solubility $\pm$ standard deviation (% w/w)
90	$49.5 \pm 0.4$	$21.4 \pm 3.0$
100	$49.7 \pm 0.0$	$23.0 \pm 0.4$
110	$50.2 \pm 0.3$	$26.1 \pm 2.3$
120	$50.8 \pm 0.1$	$30.1 \pm 0.7$
130	$51.3 \pm 0.1$	$33.4 \pm 0.7$
140	$52.4 \pm 0.2$	$40.4 \pm 1.7$



**Figure 2.** DSC traces of the second heating cycle of olanzapine–PLGA 50% w/w microparticles annealed at 70, 80, and 90 °C.



**Figure 4.** Olanzapine–PLGA solubility curve generated at temperatures between 90 and 140 °C and extrapolated using the Flory–Huggins model (solid line). The red crosses show the experimentally determined solubility, and the dashed lines represent the 95% confidence interval of olanzapine solubility at each temperature calculated by the model.



**Figure 5.** QiMTDSC method: Reversing heat capacity vs annealing time showing crystallization in the 35% and 50% w/w polymer dispersions on heating quasi-isothermally at 90 °C

generate a solubility curve (Figure 4). The experimental data were fitted with the Flory–Huggins model to calculate the interaction parameter,  $\chi$ , (and an associated error) for the system, which allowed extrapolation of the curve to any temperature by the relationship described in eq 1.  $\Delta H_{\text{fus}}$  and  $T_m$  are the heat of fusion and melting point of the crystalline drug, respectively.  $\phi_{\text{drug}}$  and  $\phi_{\text{polymer}}$  are the volume fractions of drug and polymer.  $T$  is the temperature,  $R$  is the gas constant, and  $m$  is the ratio of the volume of the polymer to that of the lattice site.

$$\frac{\Delta H_{\text{fus}}}{R} \left( \frac{1}{T_m} - \frac{1}{T} \right) = \ln(\phi_{\text{drug}}) + \left( 1 - \frac{1}{m} \right) \phi_{\text{polymer}} + \chi (\phi_{\text{polymer}})^2 \quad (1)$$

A number of studies have used this approach to estimate solubility below the  $T_g$ .<sup>9,12,15</sup>  $\chi$  was calculated as  $-3.22 \pm 0.26$

for the system, which allowed the solubility curve to be calculated at any temperature with a 95% confidence interval. Despite the reasonable solubility measured at 90 °C of 21% w/w, the model predicted it to dramatically reduce to 2% w/w at 25 °C.

**Measuring Drug–Polymer Solubility: QiMTDSC (RevC<sub>p</sub>) Demixing Method.** In the second part of this study, an alternative protocol was developed with the aim of using the same principle of heating supersaturated olanzapine–PLGA solid dispersions but using a different physical property of the solid dispersions (RevC<sub>p</sub> rather than  $T_g$ ) to measure the drug–polymer solubility. To achieve this, a QiMTDSC method was developed. An olanzapine–PLGA solid dispersion was heated and held at an elevated quasi-isothermal temperature, and as a result of the small modulation in temperature with time, the RevC<sub>p</sub> of the sample could be continually

measured. This is significant, because crystalline and amorphous forms of the same material have different heat capacities at temperatures above the  $T_g$  of the sample. Therefore, as the system undergoes demixing and olanzapine crystallizes at the elevated temperature, the total heat capacity of the solid dispersion system should reduce. The magnitude of the reduction should allow the relative fractions of amorphous and crystalline olanzapine to be calculated, which in turn will allow the solubility to be determined. Additionally, as the change in  $\text{RevC}_p$  is measured in real time and at a quasi-isothermal temperature, the kinetics of the crystallization should be able to be simultaneously measured using this protocol.

The newly developed QiMTDSC protocol requires accurate and precise  $\text{RevC}_p$  measurements. The  $\text{RevC}_p$  of the individual raw materials were measured at 90 °C. This temperature was selected, because it was the lowest temperature at which the system fully demixed (crystallized) when under the annealing conditions, which makes it most relevant to ambient storage conditions. The  $\text{RevC}_p$  of crystalline and amorphous olanzapine at the temperature of interest were found to be  $1.28 \pm 0.03$  and  $1.69 \pm 0.04$  J/g·°C, respectively. The heat capacity of amorphous PLGA was measured as  $1.87 \pm 0.04$  J/g·°C at the same temperature.

Olanzapine–PLGA solid dispersions with drug loads of 10, 35, and 50% w/w were annealed at 90 °C for 70 min (Figure 5). The 10% w/w solid dispersion was used as a negative control as no significant crystallization was expected in this system under the annealing conditions used on the basis of the solubility values calculated using the DSC demixing ( $T_g$ ) method (Table 1). The  $\text{RevC}_p$  clearly reflected this, as it remained constant at 1.86 J/g·°C throughout the experiment, after the initial equilibration period of 5 min. On the basis of the solubility measurements made from the DSC demixing ( $T_g$ ) method, the systems with 35 and 50% w/w drug load are reasonably predicted to be supersaturated, and therefore, crystallization of olanzapine was expected on annealing at 90 °C. The use of two samples with different levels of supersaturation was to assess whether the QiMTDSC protocol could distinguish between different levels of crystallization on annealing. Figure 5 shows that the protocol is capable of this, as a significantly greater reduction in  $\text{RevC}_p$  was recorded during annealing in the 50% w/w system compared to the 35% w/w system. The rank order of  $\text{RevC}_p$  following annealing was as expected on the basis of the raw material  $\text{RevC}_p$  values, with 10% w/w having the highest and 50% w/w having the lowest due to these systems having the highest and lowest amorphous content, respectively. The apparent plateau of the  $\text{RevC}_p$  signal after 20 min for the 50% w/w system was not reproducible between replicates and was attributed to fluctuations in the measurement.

**Calculation of Drug–Polymer Solubility from  $\text{RevC}_p$ .** Fractions of amorphous and crystalline olanzapine present in the solid dispersion following demixing can be calculated directly from the measured  $\text{RevC}_p$  using eq 2, providing the heat capacity of the crystalline/amorphous drug and amorphous polymer are known at the temperature of interest.

$$\text{RevC}_p^{\text{total}}(T) = \phi[x\text{RevC}_p^{\text{amorphous}}(T) + (1 - x)\text{RevC}_p^{\text{crystalline}}(T)] + (1 - \phi)\text{RevC}_p^{\text{polymer}}(T) \quad (2)$$

Qi et al. derived eq 2 as a method of calculating crystalline/amorphous fractions of drug in solid dispersion formulations.<sup>23</sup>

$\text{RevC}_p^{\text{total}}$  refers to the total heat capacity measured for the solid dispersion system,  $\text{RevC}_p^{\text{amorphous}}$  and  $\text{RevC}_p^{\text{crystalline}}$  refer to the heat capacities of the two physical forms of the drug and  $\text{RevC}_p^{\text{polymer}}$  is the heat capacity of the polymer.  $T$  in parentheses signifies the heat capacities are measured at the same temperature and are not to be used as multipliers.  $\phi$  is the weight fraction of total drug in the system, and  $x$  is the weight fraction of the amorphous form of drug.

Eq 2 assumes ideal additivity of the heat capacities, although previous studies have made an identical assumption in developing miscibility models.<sup>23,24</sup> The measured heat capacity of the 10% w/w solid dispersion provides evidence that this assumption is valid, as the heat capacity of the system is in excellent agreement with the calculated value of 1.85 J/g·°C, if the system is assumed to be fully amorphous. It should be noted, however, that eq 2 has been previously been associated with a reasonably substantial error when applied to calculate heat capacities of binary mixtures of liquids.<sup>24</sup>

Once the weight fractions of the crystalline and amorphous drug are determined using eq 2, the solubility can be calculated using eq 3.

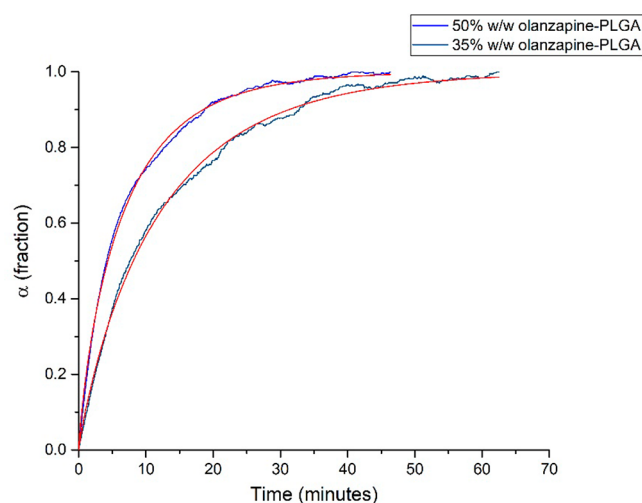
$$\text{solubility}(\%w/w) = x \cdot \phi \cdot 100 \quad (3)$$

The olanzapine–PLGA solubility at 90 °C calculated using the heat capacity data from the 35 and 50% w/w solid dispersion systems was  $23.6 \pm 6.1\%$  w/w. There was no statistically significant difference between the solubility values calculated using the two supersaturated solid dispersions (one way ANOVA,  $p = 0.58$ ); therefore, an average solubility value was calculated.

**Calculation of Crystallization Kinetics Using the QiMTDSC Method.** An advantage of the QiMTDSC method is that the  $\text{RevC}_p$  signal is effectively describing a phase change over time at a quasi-isothermal temperature, which allows the kinetics of this change to be easily calculated, despite the complexity of the system. For example, QiMTDSC has previously been used to measure crystallization kinetics in lipid-based systems.<sup>25</sup> The phase change occurring in this study is crystallization of the thermodynamic excess of olanzapine from the supersaturated molecularly dispersed state. The Avrami equation (eq 4) can be used to describe crystallization processes, whereby  $\alpha$  is the crystal fraction at time  $t$ ,  $k$  is the crystallization rate constant, and  $n$  is the Avrami exponent.<sup>26</sup> The Avrami exponent can be used to estimate the mechanism and processes involved in the crystallization event, although it can also be used as an adjustable fitting parameter that may be nonintegral.

$$\alpha = 1 - \exp(-kt^n) \quad (4)$$

The QiMTDSC data were normalized and processed so that the  $\text{RevC}_p$  was described as a fraction of the total change in the signal during the experiment (Figure 6). As a result of the time taken for equilibration at the start of the QiMTDSC experiments, data cannot be reliably collected for the first 5 min; therefore, for the purposes of the data fitting, the data collected in the first 5 min were not included, and the time at 5 min was considered  $t = 0$ . As a result of this, the first stage of the crystallization (predominantly nucleation) may not be recorded. Because of this limitation, interpretation of the calculated Avrami exponent should be treated with caution. The progress of the crystallization occurring in both solid dispersions was well-fitted with the Avrami equation ( $R^2 = \geq$



**Figure 6.** Fraction of phase change (crystallization) as a function of time for olanzapine–PLGA polymer dispersions at 90 °C (red lines show Avrami fitting).

0.997), allowing a crystallization rate constant ( $k$ ) to be calculated (Table 2).

**Table 2.** Avrami Fitting of Crystallization Data for Olanzapine Dispersions in PLGA

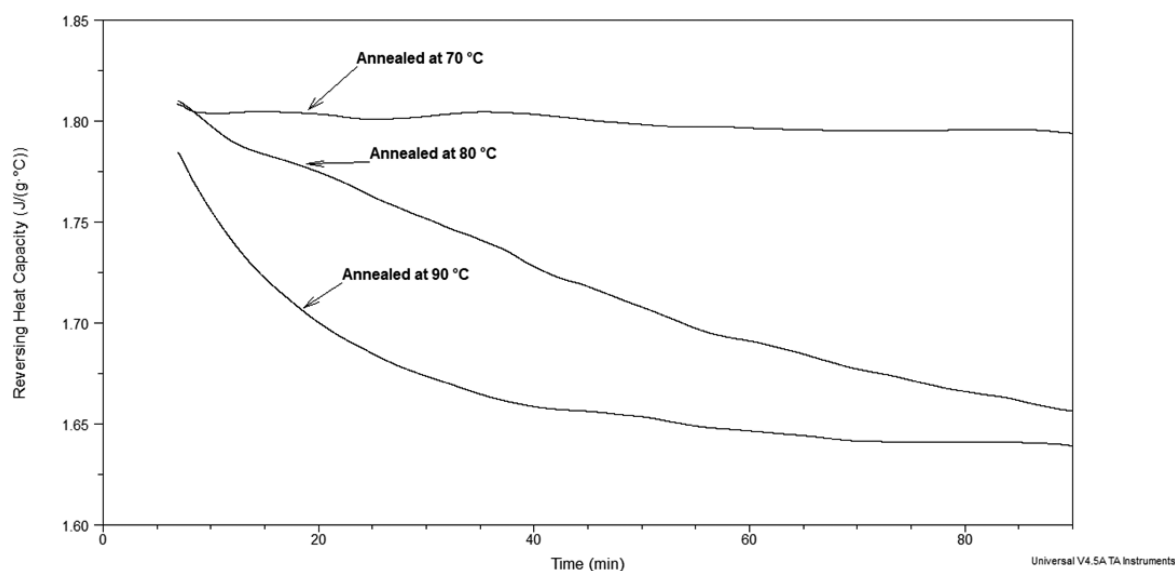
solid dispersion	$k$ ( $\text{min}^{-n}$ )	$n$	$R^2$
35% w/w OLZ/PLGA	0.108	0.890	0.997
50% w/w OLZ/PLGA	0.206	0.827	0.998

The crystallization rate constants of the two solid dispersion systems show that the 50% w/w olanzapine–PLGA system crystallizes at approximately twice the rate of the 35% w/w system at 90 °C. This demonstrates that the protocol can assess the effect of drug load on crystallization rates in solid dispersions at isothermal temperatures, which is of significance when studying the physical stability of supersaturated micro-particle solid dispersion systems.

The Avrami exponent calculated for each system was approximately 1, which suggests one-dimensional crystal growth; however, due to the limitations highlighted above, the exponent can be considered a fitting parameter to allow calculation of the rate constant, which is the parameter of interest.

The QiMTDSC method can therefore be used to assess whether crystallization (demixing) will occur at a particular temperature, and if so, how long this process will take until the equilibrium is reached. To identify the temperature range of interest, a single stepwise QiMTDSC experiment can be performed, whereby the temperature of the sample is increased in an incremental, stepwise manner, and the  $\text{RevC}_p$  is monitored to identify the lowest temperature at which crystallization occurs. For completion and comparison to the DSC demixing ( $T_g$ ) method, a 50% w/w olanzapine–PLGA solid dispersion was analyzed at 70, 80, and 90 °C, and the progression of the demixing was monitored for 90 min using the  $\text{RevC}_p$  of the sample (Figure 7). The results were consistent with the observations from the DSC ( $T_g$ ) demixing method. The  $\text{RevC}_p$  of the sample did not change on annealing at 70 °C for 90 min, showing that demixing did not occur under these conditions. However, the  $\text{RevC}_p$  steadily reduced but did not plateau when annealed for the same time at 80 °C. This demonstrates that demixing is occurring at this temperature, but the solubility equilibrium is not achieved within the time frame allowed. Annealing at 90 °C resulted in complete demixing within the time allowed, which is shown by the plateau in  $\text{RevC}_p$  after 70 min. These data are consistent with the  $T_g$  (Figure 1) and standard DSC results (Figure 2), showing that the QiMTDSC protocol is a simple way of assessing which annealing conditions are required to achieve complete demixing of a particular system.

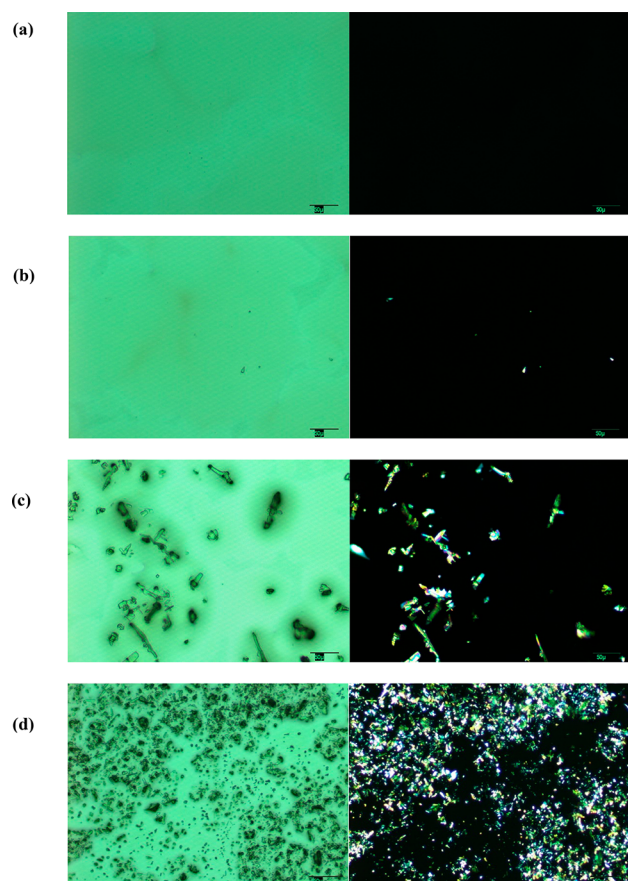
**Measuring Drug–Polymer Solubility: Demixing Microscopy Method.** A series of olanzapine–PLGA micro-particle solid dispersions with increasing drug load were evenly spread on glass slides and annealed at 90 °C for 24 h. The samples were then transferred to a hot stage set at 90 °C and analyzed using standard and polarized light microscopy to determine whether crystalline olanzapine was present in the



**Figure 7.** QiMTDSC traces showing change in  $\text{RevC}_p$  with time in 50% w/w olanzapine–PLGA at 70, 80, and 90 °C.



samples. The results of the semiquantitative microscopy method are displayed in Figure 8. The microparticles deformed



**Figure 8.** Light and polarized light microscopy images of olanzapine–PLGA solid dispersions annealed at 90 °C: (a) 10%, (b) 15%, (c) 20%, and (d) 25% w/w olanzapine load.

in shape at the elevated temperatures and produced a thin film. Figure 8a shows a solid dispersion with a 10% w/w drug load. No crystals were visible in the polymer phase, which was confirmed by the use of polarized light, suggesting that the system is unsaturated at this drug load. Figure 8b shows a solid dispersion with a 15% w/w drug load. A very small number of crystals were sparsely distributed in the polymer matrix, indicating that the system was at its saturation level at this drug load and temperature. Figure 8c,d show solid dispersions with 20 and 25% w/w olanzapine loads, respectively. Substantial olanzapine crystallites were visible, indicating that these systems are supersaturated. From these results, the olanzapine–PLGA solubility level appears to be approximately 15–20% w/w.

## DISCUSSION

**Comparison of Methods for Measuring Olanzapine–PLGA Solubility.** Three methods have been used in this study to assess the solubility of olanzapine in PLGA: (1) a DSC demixing ( $T_g$ ) method, (2) a QiMTDSC demixing ( $\text{Rev}C_p$ ) method, and (3) a hot stage microscopy demixing method. All of these methods operate on the same principle of heating supersaturated solid dispersions to promote crystallization of the thermodynamic excess of drug; however, the methods differ in how the solubility is determined and it is of interest to

see how the solubility values calculated compare, particularly as the  $\text{Rev}C_p$  method used is completely novel, the  $T_g$  method is a recent development, and no such comparison has been done previously.

Solubility is a temperature-dependent parameter; therefore, when comparing the methods, it is simpler to do so at one temperature of interest. Hence, 90 °C was selected in this study, as this was determined to be the lowest temperature at which demixing (crystallization) of the system occurred in a reasonable time frame and was therefore the solubility that was closest to ambient storage conditions that could be measured experimentally. The olanzapine–PLGA solubility values determined by each method are summarized in Table 3.

**Table 3.** Summary of Olanzapine–PLGA Solubility Determined at 90 °C Using Different Demixing Methods

method	solubility (% w/w)
DSC ( $T_g$ )	21.4 ± 3.0
QiMTDSC ( $\text{Rev}C_p$ )	23.6 ± 6.1
polarized light microscopy	15–20

The two DSC methods calculated the solubility to be very similar in the region of 21–24% w/w, which provides strong evidence that the new QiMTDSC protocol is a valid approach for measuring drug–polymer solubility. The results of the microscopy method used indicated that the system was near its saturation level at 15% w/w and supersaturated at 20% w/w. These microscopy results are only semiquantitative but suggest that the solubility level at this temperature lies at approximately 15–20% w/w, which is in reasonable agreement with the values calculated the  $T_g$  and  $\text{Rev}C_p$ , although the DSC methods do appear to slightly overcalculate solubility. The main advantage of the microscopy method is that it is not reliant on a calculation based on a theoretical relationship between a physical property of the sample and solubility; it is a definitive and intuitive method for assessing whether or not the system is supersaturated at a given temperature. A limitation of the method, however, is that it is only semiquantitative, and false positives are possible due to the high level of sensitivity; therefore, the 20% w/w system was interpreted as the first truly supersaturated system, as the level of crystallinity in the 15% w/w system was very low.

The temperature dependency of solubility means that some prediction of solubility at room temperature is of practical importance when considering the long-term physical stability of pharmaceutical systems. The Flory–Huggins model was fitted to the solubility curve generated using the DSC demixing ( $T_g$ ) method, allowing extrapolation of the curve to 25 °C (Figure 4). Olanzapine–PLGA solubility was predicted to be 2% w/w at this temperature, which is lower than drug loads typically used in PLGA microparticle formulations. This highlights that it may not always be feasible to produce thermodynamically stable systems with the required drug load, but it is important to be aware of this even if it is necessary to proceed with a supersaturated glass.

The use of the glass transition to calculate solubility is based on the principle that the  $T_g$  of a binary system can be used to predict the composition of that system using a number of different theoretical approaches, such as the Gordon–Taylor, Fox, and Couchman and Karasz equations.<sup>10,27</sup> All of these approaches make slightly different assumptions and therefore differ slightly in the prediction of the relationship between  $T_g$



and composition of the amorphous phase. One major assumption made by the Gordon–Taylor equation is that the components do not form strong specific intermolecular interactions and exhibit ideal mixing behavior. Many systems have been shown to deviate from the predicted relationship, which is suggestive of nonideal mixing behavior.<sup>28–34</sup> The system in question has been found to deviate from the predicted  $T_g$ , therefore suggesting that the system exhibits nonideal mixing behavior, which is why theoretical relationships were not used to calculate solubility in this study. Deviation from predicted values is often attributed to the presence of strong specific drug–polymer interactions, but caution should be exercised when making this inference purely from observed deviations from the Gordon–Taylor equation, as there are conflicting reports in the literature regarding this relationship.<sup>35</sup> A standard curve was therefore used to experimentally establish the relationship between  $T_g$  and olanzapine–PLGA composition. Because of these potential complications associated with quantifying the solubility from the  $T_g$  of the amorphous phase, the QiMTDSC method was investigated as an alternative approach, which uses a different physical property ( $RevC_p$ ) to calculate solubility.

**Development of QiMTDSC Method.** A new QiMTDSC protocol was developed as part of this study as an alternative method, which uses  $RevC_p$  instead of the  $T_g$  to calculate drug–polymer solubility. The  $T_g$  method does not work for systems where the drug and polymer have similar  $T_g$  values; hence, it is important to investigate alternative approaches that use alternative properties of the sample. Heat capacity is essentially a measure of molecular mobility, and amorphous materials above their  $T_g$  have significantly higher molecular mobility and therefore heat capacity than the crystalline counterpart at the same temperature. Exploitation of this heat capacity difference is the basis of this new method and is what allows the solubility to be calculated. Demixing methods have been shown to only work at temperatures  $>T_g$ ; therefore, this approach is suitable, as there should be a significant heat capacity difference between the amorphous and crystalline drug forms at temperatures where demixing occurs.<sup>9</sup> Comparison of the solubility values determined by the three demixing methods (Table 3) suggests that using  $RevC_p$  to calculate solubility is a valid approach, as it was highly comparable to that calculated using the established DSC demixing ( $T_g$ ) method. The new QiMTDSC protocol, however, appears to slightly overcalculate the drug–polymer solubility level of this system when compared to the microscopy method, which may be due in part to the difficulty of obtaining accurate and precise heat capacity measurements as very small differences in  $RevC_p$  result in significant differences in solubility calculated. This resulted in a higher standard deviation in the solubility calculations compared to the  $T_g$  method. To minimize this effect, systems that are best suited to this method will have a relatively large difference in heat capacity between the crystalline and amorphous forms of the drug of interest, and highly supersaturated systems should be used. The quality and condition of the DSC instrument and skill of the operator were also found to be important factors when making  $RevC_p$  measurements accurately enough to calculate valid solubility values; however, with future developments in DSC instruments (e.g., improved baseline quality), these measurements should become easier to make.

The main advantage of this method is that the demixing process can be monitored under different annealing conditions,

and crystallization kinetics of the drug can also be measured at a particular temperature, as the protocol is isothermal and measures the heat capacity, which changes as a function of time due to the crystallization of the drug. The crystallization kinetics can only be measured, however, if they are sufficiently slow, as the QiMTDSC protocol takes a few minutes to equilibrate, and therefore, any crystallization that occurs in this initial 5 min period cannot be recorded and analyzed.

The crystallization rate constant was calculated for olanzapine in two microparticle solid dispersion systems at 90 °C. Crystallization occurred at approximately twice the rate in the solid dispersion with a higher level of supersaturation, showing that the supersaturation level is critical and affects the rate of crystallization. Pseudoexponential crystallization kinetics were observed for both systems and have previously been reported in other drug–polymer dispersions.<sup>36</sup> These kinetics can be expected if the crystallization occurs in suitably confined geometry, which has been discussed by Descamps and Willart in a recent review article.<sup>37</sup> The microscopy images, particularly Figure 8d, support this theory and suggest that the crystallization is confined by the presence of the polymer because of the small crystallite size observed, which has interesting implications for the mechanism of polymer stabilization.

The developed protocol can therefore be used to investigate how both the composition (drug/polymer concentration) and storage conditions (temperature) affect the rate of crystallization in supersaturated microparticle solid dispersion formulations, which is important to help identify long-term physical stability issues. The protocol could therefore be used to effectively stress test systems at elevated temperatures to assess whether physical instability may be an issue for a formulation. It should be noted that the elevated temperature is likely to deform the microparticles in this case, but this allows a rapid assessment of whether crystallization of the drug may occur in a particular system, and calculation of the rate at which it occurs at various temperatures, in a dramatically accelerated time frame.

The QiMTDSC protocol was also shown to be able to easily determine whether the equilibrium solubility is achieved on demixing at a particular annealing temperature by monitoring  $RevC_p$  with time and measuring how long it takes for the  $RevC_p$  signal to plateau. This is significant, because in a single experiment, it is possible to be sure the equilibrium solubility is achieved, quantify the solubility at the annealing temperature, and model the kinetics of crystallization (demixing). As expected, the time taken to reach equilibrium solubility was shown to increase as the annealing temperature was reduced. Interestingly, however, no demixing appeared to occur at all within a 90 min time frame at 70 °C, whereas significant demixing occurred at 80 °C (Figure 7). The  $T_g$  of the system before demixing is approximately 55 °C (Figure 1); therefore, the system is above  $T_g$  at all the annealing temperatures used. This suggests that as the annealing conditions approach the  $T_g$ , the kinetics of crystallization become significantly slower to a point where no crystallization is observed at all within 90 min when this system is annealed at 15 °C above the glass transition. These data demonstrate the problem with making drug–polymer solubility measurements at ambient conditions, as the demixing kinetics become sufficiently slow, rendering measurements at temperatures lower than the  $T_g$  of the system highly impractical. Figure 7 shows that the crystallization kinetics increase as a function of temperature; however, it

should be noted that as the temperature is further increased, the rate of crystallization would eventually be expected to reduce, despite the increased molecular mobility, as the level of supersaturation and thermodynamic drive for crystallization is reduced.

In summary, the QiMTDSC protocol developed allows calculation of the thermodynamic solubility equilibrium level at elevated temperatures, direct measurement of the rates of crystallization that occur in the solid dispersion, and a simple way of assessing which conditions are required for complete demixing of the system to occur.

## CONCLUSION

A new QiMTDSC protocol was developed that has been shown to be an alternative method of measuring drug–polymer solubility. The new method was validated by comparison to an established DSC demixing ( $T_g$ ) method, and the two protocols were found to calculate very similar olanzapine–PLGA solubility levels at a temperature of interest. The two DSC methods were then compared to a simple microscopy demixing method, which identified that DSC methods may slightly overcalculate the drug–polymer solubility for this system. The main advantage of the QiMTDSC protocol was that it is also able to simultaneously measure the drug crystallization kinetics occurring within the solid dispersion and easily identify annealing conditions required to result in complete demixing of the system.

Olanzapine–PLGA microparticle formulations were the systems studied in this work. The QiMTDSC protocol has been shown to easily identify the temperature at which crystallization starts occurring in this system and has also been able to model the kinetics of this process. The microparticles may ultimately deform during this analysis, but nevertheless, the protocol provides a rapid assessment of whether physical instability may be an issue for the formulation by effectively stress testing the system with elevated temperature. Identification of potential physical instability in PLGA microparticles is of importance, as it may impact the release performance of the formulation.

## AUTHOR INFORMATION

### Corresponding Author

\*Tel: +44 20 7753 5819; Fax: +44 207 753 5560; E-mail: duncan.craig@ucl.ac.uk.

### ORCID

Simon Gaisford: 0000-0003-1000-3208

Duncan Q. M. Craig: 0000-0003-1294-8993

### Author Contributions

The manuscript was written through contributions of all authors. All authors have given approval to the final version of the manuscript.

### Notes

The authors declare no competing financial interest.

## ACKNOWLEDGMENTS

The authors would like to thank and acknowledge the Centre for Doctoral Training in Advanced Therapeutics and Nanomedicine and the EPSRC for funding (Grant No. EP/L01646X). The authors would also like to acknowledge Dr. Matthias Knopp for his assistance applying the Flory–Huggins model to the experimental solubility data.

## ABBREVIATIONS

DCM, dichloromethane; DSC, differential scanning calorimetry; PC-SAFT, perturbed-chain statistical associating fluid theory; PLGA, poly(lactic-co-glycolic acid); PLM, polarized light microscopy; QiMTDSC, quasi-isothermal modulated temperature differential scanning calorimetry;  $RevC_p$ , reversing heat capacity;  $T_g$ , glass transition temperature

## REFERENCES

- (1) Vo, C. L.-N.; Park, C.; Lee, B.-J. Current Trends and Future Perspectives of Solid Dispersions Containing Poorly Water-Soluble Drugs. *Eur. J. Pharm. Biopharm.* **2013**, *85* (3), 799–813.
- (2) Marsac, P. J.; Shamblin, S. L.; Taylor, L. S. Theoretical and Practical Approaches for Prediction of Drug–Polymer Miscibility and Solubility. *Pharm. Res.* **2006**, *23* (10), 2417–2426.
- (3) Grohgan, H.; Priemel, P. a.; Löbmann, K.; Nielsen, L. H.; Laitinen, R.; Mullertz, A.; Van den Mooter, G.; Rades, T. Refining Stability and Dissolution Rate of Amorphous Drug Formulations. *Expert Opin. Drug Delivery* **2014**, *11* (6), 977–989.
- (4) Prudic, A.; Ji, Y.; Sadowski, G. Thermodynamic Phase Behavior of API/Polymer Solid Dispersions. *Mol. Pharmaceutics* **2014**, *11*, 2294–2304.
- (5) Prudic, A.; Lesniak, A. K.; Ji, Y.; Sadowski, G. Thermodynamic Phase Behaviour of Indomethacin/PLGA Formulations. *Eur. J. Pharm. Biopharm.* **2015**, *93*, 88–94.
- (6) Luebbert, C.; Huxoll, F.; Sadowski, G. Amorphous–Amorphous Phase Separation in API/Polymer Formulations. *Molecules* **2017**, *22* (2), 296.
- (7) Tao, J.; Sun, Y.; Zhang, G. G. Z.; Yu, L. Solubility of Small-Molecule Crystals in Polymers: D-Mannitol in PVP, Indomethacin in PVP/VA, and Nifedipine in PVP/VA. *Pharm. Res.* **2009**, *26* (4), 855–864.
- (8) Sun, Y. E.; Tao, J.; Zhang, G. G. Z.; Yu, L. Solubilities of Crystalline Drugs in Polymers: An Improved Analytical Method and Comparison of Solubilities of Indomethacin and Nifedipine in PVP, PVP/VA, and PVAc. *J. Pharm. Sci.* **2010**, *99* (9), 4023–4031.
- (9) Mahieu, A.; Willart, J. F.; Dudognon, E.; Danede, F.; Descamps, M. A New Protocol to Determine the Solubility of Drugs into Polymer Matrices. *Mol. Pharmaceutics* **2013**, *10* (2), 560–566.
- (10) Gordon, M.; Taylor, J. S. Ideal Copolymers and the Second-Order Transitions of Synthetic Rubbers. i. Non-Crystalline Copolymers. *J. Appl. Chem.* **1952**, *2* (9), 493–500.
- (11) Knopp, M. M.; Olesen, N. E.; Holm, P.; Löbmann, K.; Holm, R.; Langguth, P.; Rades, T. Evaluation of Drug–Polymer Solubility Curves Through Formal Statistical Analysis: Comparison of Preparation Techniques. *J. Pharm. Sci.* **2015**, *104* (1), 44–51.
- (12) Knopp, M. M.; Olesen, N. E.; Holm, P.; Langguth, P.; Holm, R.; Rades, T. Influence of Polymer Molecular Weight on Drug–polymer Solubility: A Comparison between Experimentally Determined Solubility in PVP and Prediction Derived from Solubility in Monomer. *J. Pharm. Sci.* **2015**, *104* (9), 2905–2912.
- (13) Knopp, M. M.; Olesen, N. E.; Huang, Y.; Holm, R.; Rades, T. Statistical Analysis of a Method to Predict Drug–Polymer Miscibility. *J. Pharm. Sci.* **2016**, *105* (1), 362–367.
- (14) Knopp, M. M.; Tajber, L.; Tian, Y.; Olesen, N. E.; Jones, D. S.; Kozyra, A.; Löbmann, K.; Paluch, K.; Brennan, C. M.; Holm, R.; Healy, A. M.; Andrews, G. P.; Rades, T. Comparative Study of Different Methods for the Prediction of Drug–Polymer Solubility. *Mol. Pharmaceutics* **2015**, *12* (9), 3408–3419.
- (15) Wlodarski, K.; Sawicki, W.; Kozyra, A.; Tajber, L. Physical Stability of Solid Dispersions with Respect to Thermodynamic Solubility of Tadalafil in PVP-VA. *Eur. J. Pharm. Biopharm.* **2015**, *96*, 237–246.
- (16) Matsuo, T. Low Temperature Thermal Properties of Amorphous Materials. *Pure Appl. Chem.* **1998**, *70* (3), 599–602.
- (17) Kane, J. M.; Detke, H. C.; Naber, D.; Sethuraman, G.; Lin, D. Y.; Bergstrom, R. F.; McDonnell, D. Olanzapine Long-Acting Injection: A 24-Week, Randomized, Double-Blind Trial of Main-

tenance Treatment in Patients with Schizophrenia. *Am. J. Psychiatry* **2010**, 167 (2), 181–189.

(18) Kapoor, D. N.; Bhatia, A.; Kaur, R.; Sharma, R.; Kaur, G.; Dhawan, S. PLGA: A Unique Polymer for Drug Delivery. *Ther. Delivery* **2015**, 6 (1), 41–58.

(19) Kaplan, G.; Casoy, J.; Zummo, J. Impact of Long-Acting Injectable Antipsychotics on Medication Adherence and Clinical, Functional, and Economic Outcomes of Schizophrenia. *Patient Prefer. Adherence* **2013**, 7, 1171–1180.

(20) Enriquez, G. G.; Rizvi, S. A. A.; D'Souza, M. J.; Do, D. P. Formulation and Evaluation of Drug-Loaded Targeted Magnetic Microspheres for Cancer Therapy. *Int. J. Nanomed.* **2013**, 8 (1), 1393–1402.

(21) Nahata, T.; Saini, T. R. Optimization of Formulation Variables for the Development of Long Acting Microsphere Based Depot Injection of Olanzapine. *J. Microencapsulation* **2008**, 25 (6), 426–433.

(22) Seju, U.; Kumar, A.; Sawant, K. K. Development and Evaluation of Olanzapine-Loaded PLGA Nanoparticles for Nose-to-Brain Delivery: In Vitro and in Vivo Studies. *Acta Biomater.* **2011**, 7 (12), 4169–4176.

(23) Qi, S.; Belton, P.; Nollenberger, K.; Clayden, N.; Reading, M.; Craig, D. Q. M. Characterisation and Prediction of Phase Separation in Hot-Melt Extruded Solid Dispersions: A Thermal, Microscopic and NMR Relaxometry Study. *Pharm. Res.* **2010**, 27 (9), 1869–1883.

(24) Teja, A. S. Simple Method for the Calculation of Heat Capacities of Liquid Mixtures. *J. Chem. Eng. Data* **1983**, 28 (1), 83–85.

(25) Otun, S. O.; Meehan, E.; Qi, S.; Craig, D. Q. M. The Use of Quasi-Isothermal Modulated Temperature Differential Scanning Calorimetry for the Characterization of Slow Crystallization Processes in Lipid-Based Solid Self-Emulsifying Systems. *Pharm. Res.* **2015**, 32 (4), 1316–1324.

(26) Avrami, M. Kinetics of Phase Change. I General Theory. *J. Chem. Phys.* **1939**, 7 (12), 1103–1112.

(27) Couchman, P. R.; Karasz, F. E. A Classical Thermodynamic Discussion of the Effect of Composition on Glass-Transition Temperatures. *Macromolecules* **1978**, 11 (1), 117–119.

(28) Gupta, P.; Bansal, A. K. Molecular Interactions in Celecoxib-PVP-Meglumine Amorphous System. *J. Pharm. Pharmacol.* **2005**, 57 (3), 303–310.

(29) Nair, R.; Nyamweya, N.; Gönen, S.; Martínez-Miranda, L. J.; Hoag, S. W. Influence of Various Drugs on the Glass Transition Temperature of Poly(Vinylpyrrolidone): A Thermodynamic and Spectroscopic Investigation. *Int. J. Pharm.* **2001**, 225 (1–2), 83–96.

(30) Gashi, Z.; Censi, R.; Malaj, L.; Gobetto, R.; Mozzicafreddo, M.; Angeletti, M.; Masic, A.; Di Martino, P. Differences in the Interaction between Aryl Propionic Acid Derivatives and Poly(Vinylpyrrolidone) K30: A Multi-Methodological Approach. *J. Pharm. Sci.* **2009**, 98 (3), 4216–4228.

(31) Andrews, G. P.; Abudiak, O. A.; Jones, D. S. Physicochemical Characterization of Hot Melt Extruded Bicalutamide- Polyvinylpyrrolidone Solid Dispersions. *J. Pharm. Sci.* **2010**, 99 (3), 1322–1335.

(32) Tong, P.; Zografi, G. A Study of Amorphous Molecular Dispersions of Indomethacin and Its Sodium Salt. *J. Pharm. Sci.* **2001**, 90 (12), 1991–2004.

(33) Shamblin, S. L.; Huang, E. Y.; Zografi, G. The Effects of Colyophilized Polymeric Additives on the Glass Transition Temperature and Crystallization of Amorphous Sucrose. *J. Therm. Anal.* **1996**, 47, 1567–1579.

(34) Shamblin, S. L.; Taylor, L. S.; Zografi, G. Mixing Behavior of Colyophilized Binary Systems. *J. Pharm. Sci.* **1998**, 87 (6), 694–701.

(35) Baird, J. A.; Taylor, L. S. Evaluation of Amorphous Solid Dispersion Properties Using Thermal Analysis Techniques. *Adv. Drug Delivery Rev.* **2012**, 64 (5), 396–421.

(36) Hédoux, A.; Guinet, Y.; Descamps, M. The Contribution of Raman Spectroscopy to the Analysis of Phase Transformations in Pharmaceutical Compounds. *Int. J. Pharm.* **2011**, 417 (1–2), 17–31.

(37) Descamps, M.; Willart, J. F. Scaling Laws and Size Effects for Amorphous Crystallization Kinetics: Constraints Imposed by

Nucleation and Growth Specificities. *Int. J. Pharm.* **2018**, 542 (1–2), 186–195.