Assessment of Crystallinity in Processed Sucrose by Near-Infrared Spectroscopy and Application to Lyophiles

PAUL E. LUNER,[1,2] JEFFERY J. SEYER[1,3]

[1]College of Pharmacy, Division of Pharmaceutics, The University of Iowa, Iowa City, Iowa 52242

[2]Current Affiliation: Boehringer Ingelheim Pharmaceuticals Inc, Pharmaceutical Research and Development, Ridgefield, Connecticut 06877

[3]Current Affiliation: Pfizer Consumer Healthcare, Richmond, Virginia 23261

ABSTRACT: The objective of this study was to investigate the capability of near-infrared spectroscopy (NIRS) to determine crystallinity in processed sucrose using a common set of calibration standards derived from binary physical mixtures. NIRS was applied as a primary method using binary mixtures of amorphous and crystalline standards to predict crystallinity in sucrose that was either rendered partially amorphous by milling, partially recrystallized from the amorphous phase, or amorphous lyophiles annealed to induce recrystallization. Crystallinity prediction in the case of milled crystalline and recrystallized amorphous sucrose was feasible using the two-state binary calibration mixtures applying a univariate model. NIRS results for milled sucrose were comparable to those obtained using X-ray powder diffraction. The changes in crystallinity after milling and recrystallization showed expected trends. However, the same NIRS univariate calibration method could not be successfully applied for directly through the vial. To overcome this complication, NIRS was applied as a secondary method relative to water vapor sorption (WVS) where a set of processed samples were measured using both NIRS and WVS and a partial least-squares model applied. The NIRS secondary method was successfully applied and provided a standard error of calibration of 2.11% and standard error of prediction of 3.76%. © 2014 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 103:2884–2895, 2014

Keywords: amorphous; crystallinity; moisture sorption; near-infrared spectroscopy; milling; lyophilization; degree of crystallinity; diffuse reflectance; crystallization; water sorption

INTRODUCTION

A wide variety of solid-state analytical techniques have been used to determine crystallinity of pharmaceutical materials[1–4] and this remains an important area of research as amorphous forms of therapeutic agents become used more frequently to overcome solubility and dissolution limitations. Solid-state characterization methods used to determine crystallinity or amorphous content typically involve development of a calibration using physical mixtures prepared from "100%" amorphous and "100%" crystalline reference materials. In many reports, the validation is conducted using "unknowns" that are themselves prepared from physical mixtures.[5–9] In the case of milling, processed samples will likely contain a continuum of amorphous and crystalline regions throughout a given particle[10] and a more rigorous test of the suitability of a method for "real" samples involves determination in processed samples that represent true unknowns.[11–15] The concept of a one-state model[16] for crystallinity was investigated by Suryanarayanan and Mitchell[17] and more recently has been further examined.[10,18] The incorporation of this concept in method development for solid-state determinations, particularly in the case of amorphous content may have a significant bearing on the success of the method.[10] This is because the development of an accurate and robust method for crystallinity determina-

tion will depend on the extent to which the calibration samples represent the analyte and the extent to which the analytical method is sensitive to the property being measured, as highlighted by Shalaev and Zografi.[19] We have previously demonstrated the utility of near-infrared spectroscopy (NIRS) as a primary method for the determination of crystallinity using binary physical mixture of crystalline and amorphous sucrose as well as indomethacin,[20] and its general theory and application to pharmaceutical systems have been reviewed.[21,22] NIRS has been applied by other investigators to compounds for crystallinity and other solid-state phase determinations using a variety of data analysis approaches[4,14,23–26] and Bai et al.[23] have applied NIRS for determining crystallinity of lyophilized glycine. Therefore, an initial objective of this work was to extend the application of NIRS to determine crystallinity to a range of processed samples.

Pharmaceutical materials can be amorphized in a variety of ways,[27,28] and recent research has shown that amorphous materials prepared by different methods may have structural differences.[19] Consequently, solid-state analytical calibration models using reference materials that do not accurately represent the structural or thermal history of the processed amorphous material could lead to erroneous or biased results when conducting measurements on processed samples. Because NIRS is sensitive to solid-state structural features of compounds, including hydrogen bonding and intermolecular interactions that influence vibrational behavior,[3,29,30] it has been increasingly applied as a solid-state characterization method, but it is also this same sensitivity that may interfere with the interchangeability of calibrants relative to processed unknown

samples. To better understand these potential limitations in the context of NIRS analysis, we examined the question of whether a two-state calibration model constructed from binary physical mixtures can be applied successfully to determine crystallinity in processed materials that are semicrystalline in nature and represent one-state systems that have a continuum of structure ranging from amorphous to crystalline. We have used sucrose as a model compound and tested whether NIRS developed as a primary method using a specific set of binary mixtures could be applied to predict crystallinity in (1) crystalline material that was rendered partially amorphous by milling, (2) material partially recrystallized from the amorphous phase, and (3) lyophilized (amorphous) samples annealed to cause partial recrystallization. Sucrose was chosen as a model compound because it can be rendered amorphous by a variety of methods, can be crystallized under controlled conditions, and the crystalline phase has no polymorphs at room temperature and pressure. We report that crystallinity prediction in the case of milled and recrystallized sucrose was possible using the two-state binary calibration mixtures applying a simple univariate model. In the case of sucrose lyophiles, the two-state calibration model could not be successfully applied for direct interrogation through the base of the vial. To overcome this complication, NIRS was applied as a secondary method relative to water vapor sorption (WVS) as the primary method for crystallinity determination using a multivariate model. NIRS then provided an accurate representation of sample crystallinity subsequent to initial calibration when applied in this mode for determination of crystallinity in lyophiles determined directly through the vial. To our knowledge, the use of NIRS in this manner for crystallinity determination in lyophiles has not been previously reported. The applicability of the two-state calibration model (or lack thereof) in the experimental systems tested is discussed in the context of the amorphous phase and its influence on the specific spectral regions used for the quantitative model.

EXPERIMENTAL

Sucrose (99.5%) was obtained from Sigma Chemical Company (St. Louis, Missouri). The preparation of reference crystalline and reference amorphous sucrose was performed using the procedure reported by Seyer et al.,[20] wherein amorphous standard material was prepared by quenching an aqueous sucrose solution in liquid nitrogen followed by lyophilization. Particle size was controlled by sieving the crystalline and amorphous sucrose and using the fraction between 38 and 106 μm. The crystalline or amorphous character of the reference materials was confirmed by X-ray powder diffraction (XRPD) (Fig. 1). An 11-sample set of physical mixtures of amorphous and crystalline sucrose reference materials were prepared over the range of 0%–100% crystalline using an established procedure[20] and used to generate the initial binary mixture calibration model. Samples were mixed for over 30 s on a vortex mixer before acquiring each replicate spectrum to allow for different presentation to the instrument. This mixing process was previously shown to be adequate for obtaining uniformity of crystalline/amorphous sucrose physical mixtures.[20]

Milled crystalline sucrose was prepared from the crystalline reference material using a Fritsch planetary (ball) micro mill (Pulverisette 7; Fritsch, Idar-Oberstein, Germany) by milling uniform two-gram charges that were loaded in the milling
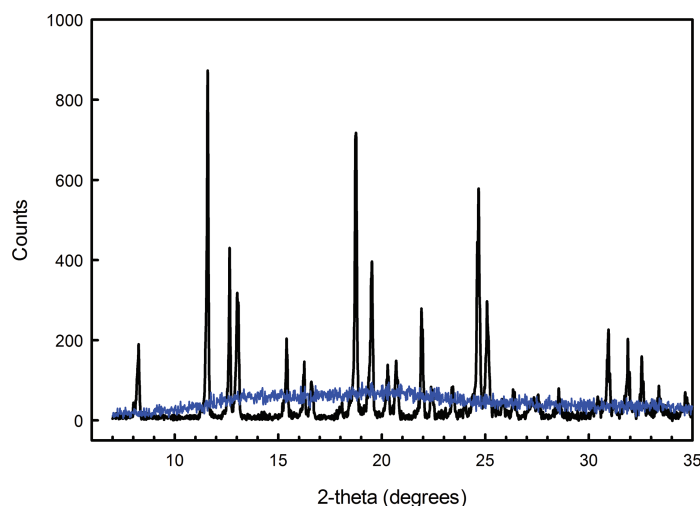

Figure 1. X-ray powder diffraction scans for crystalline (black) and amorphous sucrose (blue) reference standard materials used in the binary calibration mixtures.

chamber in a glove box (<5% relative humidity). The milling chamber was enclosed in three tightly sealed plastic bags to prevent the uptake of moisture during milling. Samples were milled for varying lengths of time and transferred to glass vials in the glove box. To prepare partially crystalline samples from the amorphous phase, samples of sucrose reference amorphous were placed in petri dishes and allowed to crystallize in a drying oven (Isotemp Model 725G; Fisher Scientific, Itasca, Illinois) at 85°C. Samples were removed from the drying oven after varying lengths of time, lightly triturated, and transferred into glass vials in a glove box at less than 5% relative humidity.

Lyophilized sucrose cakes were prepared using a 10% aqueous sucrose solution (w/v) that was pipetted (12 mL) into lyophilization vials (30 mL capacity). These samples were then lyophilized in an AdVantage Freeze Dryer (VirTis; Gardiner, New York) with a primary cycle of −46°C for 3 days, −34°C for 1 day, 0°C for 1 day followed by secondary drying at 25°C for 1 day followed by 1 day at 40 °C. Vials were then removed and stoppered, capped in a glove box (<5% relative humidity) and stored in desiccators over anhydrous calcium sulfate. Samples were removed periodically and heated in a drying oven at 65°C to induce partial crystallization. Heating times varied from 0 to 4 h. Samples were allowed to cool and near-infrared (NIR) spectra were acquired for each sample. When additional WVS testing was conducted, each sample was gently powdered in a glove box, placed in a sealed glass vial, and stored in a freezer at −30°C until used for measurement.

Water vapor sorption analysis was performed on the binary crystalline/amorphous reference mixtures and the partially recrystallized lyophiles using a VTI SGA-100 Symmetrical Gravimetric Analyzer (VTI Corporation, Hialeah, Florida) equipped with a chilled mirror dew point analyzer (Model 2000 Dewprime DF; Edgetech, Milford, Massachusetts). The instrument was equipped with ultrahigh purity dry nitrogen gas. Each run included an initial drying phase in which the material was heated to 35°C and purged with dry nitrogen at a relative humidity of less than 1%. The experimental preparation for standards and samples included an initial drying phase under less than 1%