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Tunable Diode Laser Absorption Spectroscopy (TDLAS) Enabled SMART Freeze-Dryer Technology

Gong, E. S.[a*]; Khakpash, N.[b]; Hinds, M. [a]; Schomber, L.[c]; Yusoff, Z.[c]; Pikal, M.[b]; Kessler, W.[a]

[a] Physical Sciences Inc., Andover, Massachusetts, 01810, USA
[b] Department of Pharmaceutical Sciences, School of Pharmacy, University of Connecticut, Storrs, Connecticut, 06269, USA
[c] SP Scientific, Warminster, Pennsylvania, 18974, USA

*E-mail of the corresponding author: egong@psicorp.com

Abstract

Tunable Diode Laser Absorption Spectroscopy (TDLAS) measurements combined with SMART Freeze-Dryer Technology enabled automated pharmaceutical lyophilization process development. The sensor measured water vapor temperature, density and flow velocity used to calculate water vapor mass flow rates (dm/dt). The dm/dt values were combined with a heat and mass transfer model of lyophilization to enable real-time determinations of product temperature during sublimation. The temperatures combined with a process development algorithm produced efficient freeze-drying cycles during a single lyophilization experiment. The SMART-TDLAS Freeze-Dryer Technology was used to dry placebo formulations in laboratory and pilot-scale lyophilizers, demonstrating application at multiple scales.

Keywords: *TDLAS; SMART Freeze-drying; Process development; SMART-MTM; SMART-TDLAS*

1. Introduction

Many pharmaceutical products, most notably biopharmaceuticals, must be lyophilized, or freeze-dried, to meet stability requirements for a commercializable product. During the lyophilization process, frozen water is removed through sublimation at low temperatures and pressures. [1] This leaves behind a dried product that is stable and can be stored and reconstituted at the time of patient use. The process must be designed to avoid the frozen product exceeding the formulation dependent critical temperature above which there will be viscous flow and adverse effects on the dried structure that lead to increased residual moisture and reconstitution times as well as poor stability. [1] However, processes that are overly conservative can add significant process time resulting in a non-economical manufacturing process. Generally, reducing the product temperature by 1°C will increase the primary drying time, the longest portion of the process, by 13%. [2] Freeze-drying cycles are often days long, and non-optimized cycles can drastically increase production costs and lower processing capacity.

The rapid growth of biopharmaceuticals including injectable solutions, proteins, peptides, cell and gene therapies, and vaccines has resulted in increased demand for freeze-drying. However, there is limited worldwide expertise in freeze-drying process design. The SMART Freeze-Dryer algorithm was developed by lyophilization experts at the University of Connecticut and Purdue University to enable rapid process design, even for novice users. The SMART technology requires user defined input parameters for product and vial characteristics and in-process product temperature determinations to automatically develop a freeze-drying cycle. The algorithm incorporates a pseudo-steady state heat and mass transfer model of freeze-drying in vials and expert knowledge to determine appropriate shelf temperature and process pressure set points. [3]

The SMART algorithm requires a process analytical technology (PAT) tool to determine the product temperature during the freeze-drying process. The original algorithm depended upon Manometric Temperature Measurement (MTM) technology based on pressure rise measurements to determine the product temperature. [3] Using this method, the isolation valve between the lyophilizer chamber and condenser is quickly closed and the chamber pressure (P_c) rise is measured for approximately 25 seconds. The pressure in the chamber rises due to on-going sublimation, and the pressure rise is fit to an equation to determine the vapor pressure of ice at the sublimation interface (P_{ice}). P_{ice} is used to calculate the product temperature. [3] There are limitations to the MTM approach for calculating product temperature: 1) Requires a fast-closing valve (<1s) which is infeasible for larger pilot and production-scale dryers, 2) Product temperature measurement accuracy diminishes during the cycle due to reduced sublimation caused by increased product resistance to drying (Rp) associated with a thickening dry layer, and 3) Some products, mainly high weight percent amorphous products, reabsorb water in the dried layer during the pressure rise test, leading to a reduced pressure rise and incorrect product temperature determination. [4]

The limitations of MTM motivated this work focusing on the application of an alternate approach for determining product temperature, Tunable Diode Laser Absorption Spectroscopy (TDLAS). The TDLAS sensor measures water vapor absorption lineshapes at 1392.5 nm at two measurement angles (45 and 135 degrees) with respect to the vapor flow

axis in the spool connecting the drying chamber and the condenser as shown in Fig. 1. [5] From the measured lineshapes, the gas temperature (K) can be determined using the full width at half maximum. The gas temperature is then used to calculate the absorption linestrength, S(T). Using the Beer-Lambert law, the linestrength and the integrated area under the absorption lineshape are used to determine the water vapor concentration or density (molecules cm^{-3}). The gas flow velocity (m/s) in the spool connecting the lyophilizer chamber and condenser is determined using the frequency or wavelength Doppler shift between the lineshapes from the two line-of-sight measurement locations. The gas concentration, velocity and spool cross-sectional area are used to calculate the water vapor mass flow rate (g/s). The average rate is determined by additional real-time calculation of the flow development within the duct, assuming an axi-symmetric flow profile, to apply scaling factors to the line-of-sight measurements to account for the variable gas temperature, density and velocity as a function of radial position within the duct.. The TDLAS sensor can be implemented on any scale freeze-dryer, and its accuracy is not affected by the product. Use of the TDLAS sensor to determine product temperature in the SMART Freeze-Dryer eliminates the limitations of MTM and increases the applicability of SMART Dreeze-Dryer algorithm.

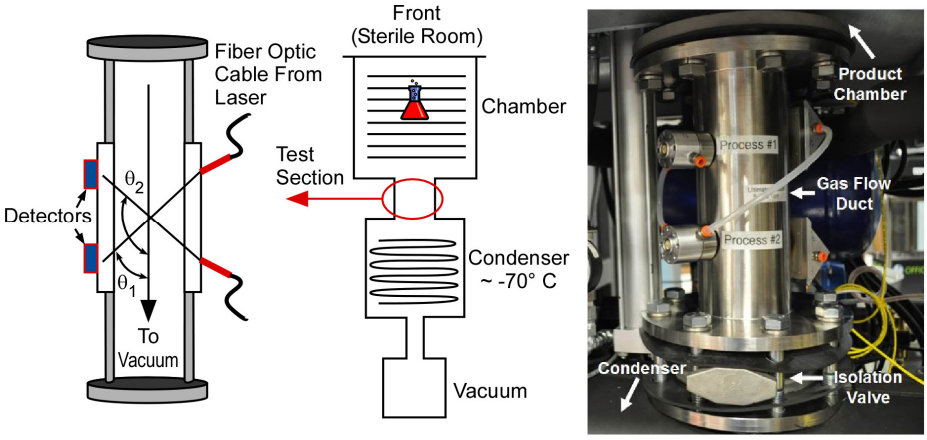

Fig. 1. Schematic diagram and photograph of the TDLAS measurement configuration within a laboratory scale lyophilizer outfitted with a two line-of-sight optical spool.

2. Materials and Methods

2.1. Materials

Mannitol, sucrose, trehalose, glycine and polyvinylpyrrolidone (PVP) were purchased from MilliporeSigma (St. Louis, MO). Solutions were prepared using ultrapure deionized water and filtered with a 0.22 µm polyethersulfone filter (Thermo Fisher Scientific, Waltham, MA). Bovine serum albumin (BSA) was purchased from MilliporeSigma (St. Louis, MO). BSA solutions were also prepared using ultrapure deionized water. Two rounds of dialysis totaling 24 hours were performed using a cellulose membrane with a molecular weight cut off of 6-8 kDa (Spectrum Chemicals, Brunswick, NJ) to remove salts. The final BSA concentration was determined using UV slope spectroscopy (C Technologies, Bridgewater, NJ). An extinction coefficient of 0.647 was used for BSA. Dialyzed BSA solutions were diluted to

the desired final concentration and filtered using a 0.22 µm syringe filter prior to use. Vials used were 20cc glass tubing vials (either Schott, Lebanon, PA or Amcor, now Nippon Electric Glass, Shiga, Japan).

2.2. Equipment

Laboratory-scale experiments were conducted using a LyoStar 3 freeze-dryer (SP Scientific, Gardiner, NY) containing three shelves with a total shelf surface area of 0.43 m^2. Pilot-scale experiments were conducted in a LyoConstellation S20 (SP Scientific, Gardiner, NY) freeze-dryer containing five shelves with a total shelf surface area of 1.86 m^2. Both systems were equipped with a LyoFlux® (Physical Sciences Inc., Andover, MA) TDLAS sensor, the LyoS™ Software Control System (SP Scientific, Gardiner, NY) and the existing SMART MTM-based Freeze-Dryer technology, as well as the custom SMART TDLAS-based software. The existing TDLAS sensor and freeze dryer software were configured to enable communication and data transfer between the two systems. The TDLAS sensor computer was connected by an Ethernet cable to the programmable logic controller (PLC) of the freeze-dryer via a separate network interface. The inter-machine communication was used to transfer status updates, alarms and system parameters between the freeze-dryer and the TDLAS sensor data and freeze-dryer shelf set points.

2.3. Experimental Setup

Product temperatures were measured in a representative subset of vials using 36 gauge bare lead thermocouple (TC) probes (Omega, Norwalk, CT) inserted into a standard lyophilization stopper and placed at the bottom center of the vial. Vial heat transfer coefficients (K_v) were experimentally determined by subliming deionized water under steady state conditions. and calculated using Equation 1:

$$K_v = \left(\Delta H_s * {dm}/{dt}\right)/\left(A_v(T_s - T_b)\right) \quad (1)$$

where ΔH_s is the heat of sublimation of ice, dm/dt is the water vapor mass flow rate, A_v is the area of the bottom of the vial, T_s is the shelf temperature and T_b is the product temperature at the bottom center of the vial. A_v was taken from engineering data for each vial type. The water vapor mass flow rates (dm/dt) during these experiments were determined using both TDLAS and gravimetric measurements. T_s and T_b were measured using temperature probes. A heat of sublimation for ice of 670 cal/g was used.

3. Results and Discussion

3.1. TDLAS product temperature calculation accuracy

The SMART algorithm requires input of the product temperature to determine shelf temperature set points required to maintain the product temperature below the product formulation critical temperature (collapse temperature, T_c, for amorphous formulations and eutectic melt, T_{eu}, for crystalline formulations). The SMART TDLAS-based Freeze-Dryer technology utilizes batch average water vapor mass flow rate values measured by the TDLAS sensor in conjunction with the pseudo steady-state heat and mass transfer model of freeze-drying to calculate product temperatures in real-time during freeze drying. The accuracy of

the calculated product temperature is dependent on the accuracy of the measured water vapor mass flow rate using the TDLAS sensor as well as the accuracy of the vial heat transfer coefficient used as a model input. Water vapor mass flow rate measurement accuracy was assessed using comparative experiments between TDLAS and gravimetric measurements of the integrated mass of water removed during ice slab experiments. In these experiments, ice slabs containing a known weight of water are formed in plastic lined frames placed directly on the lyophilizer shelves. Following sublimation experiments removing approximately 30% of the water used to form the ice slabs, comparisons of integrated amounts of water removed indicate that the TDLAS measurements generally had +/-5% error compared to gravimetric determinations with all measurements within +/-7%.

Product temperature measurement accuracy was assessed during both SMART and fixed-recipe freeze-drying cycles. The product temperature at the bottom center of a representative set of selected vials was measured using thermocouples and the batch average product temperature was calculated using the TDLAS mass flow measurements. In addition, during these experiments MTM was used to determine the product temperature at the bottom of the vial.

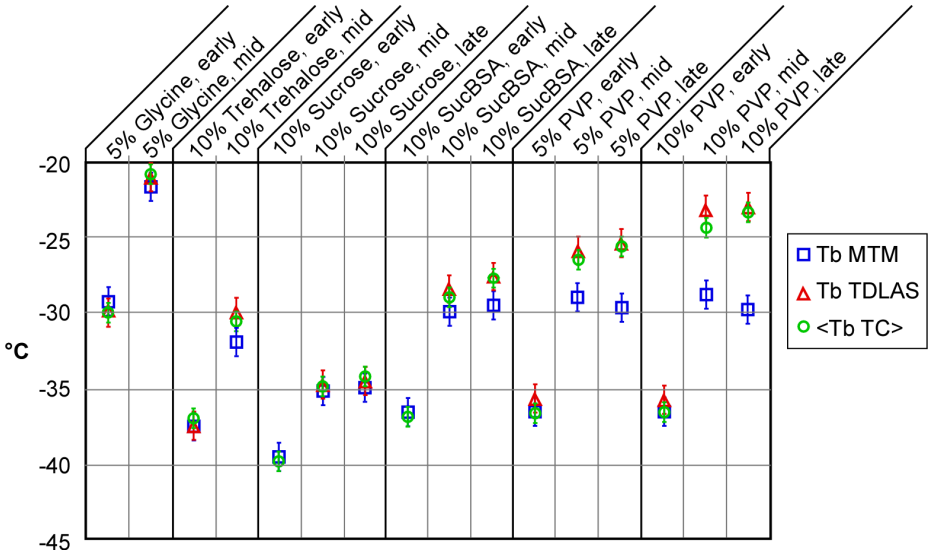

Fig. 2 Comparison of thermocouple, MTM and TDLAS measurements of the product temperature at the bottom of the vial undergoing freeze drying. Error bars: +1°C for MTM & TDLAS; 0.8°C for thermocouples.

Fig. 2 shows a comparison of these three measurement techniques for a range of product formulations. A comparison between the TDLAS product temperature and the thermocouple "gold standard" shows very good agreement (within ±1°C) between the two techniques for all formulations. A comparison with the MTM technique shows that for some formulations MTM has good agreement, but for some amorphous formulations (e.g. trehalose, sucrose/BSA and PVP) the MTM technique has unacceptable error, especially mid to late in the cycle.

3.2. SMART freeze drying of placebo formulations

Placebo formulations were used to test the SMART TDLAS-based Freeze-Dryer with relevant formulations. Sucrose is a common stabilizer and forms an amorphous product with a low collapse temperature (-32°C). [6] Mannitol is a common bulking agent and forms a crystalline product with a high eutectic melt temperature that is dependent on the crystalline structure. [7] A formulation of BSA and sucrose was used as a model for a protein formulation. A high concentration, 20% total solids with 1:1 BSA to sucrose, was used to demonstrate unsuccessful MTM measurements and cycle development compared to the successful TDLAS measurements and control. Selected experiments were conducted in both a lab-scale freeze dryer (LyoStar 3) and a pilot-scale freeze dryer (LyoConstellation S20).

Fig. 3 shows the full freeze drying cycle generated by the SMART TDLAS-based Freeze-Dryer for 20% w/w sucrose. During the the freezing and secondary drying process, the software determines the cycle parameters based on the nature of the drug product, amorphous or crystalline, and the fill depth. Freezing steps may be altered by the user based on the thermal response of the product (e.g. the addition of an annealing step). The chamber pressure set point during primary and secondary drying was determined from the target product temperature, and the shelf temperature set points during primary drying were determined by the algorithm based on TDLAS-determined product temperatures.

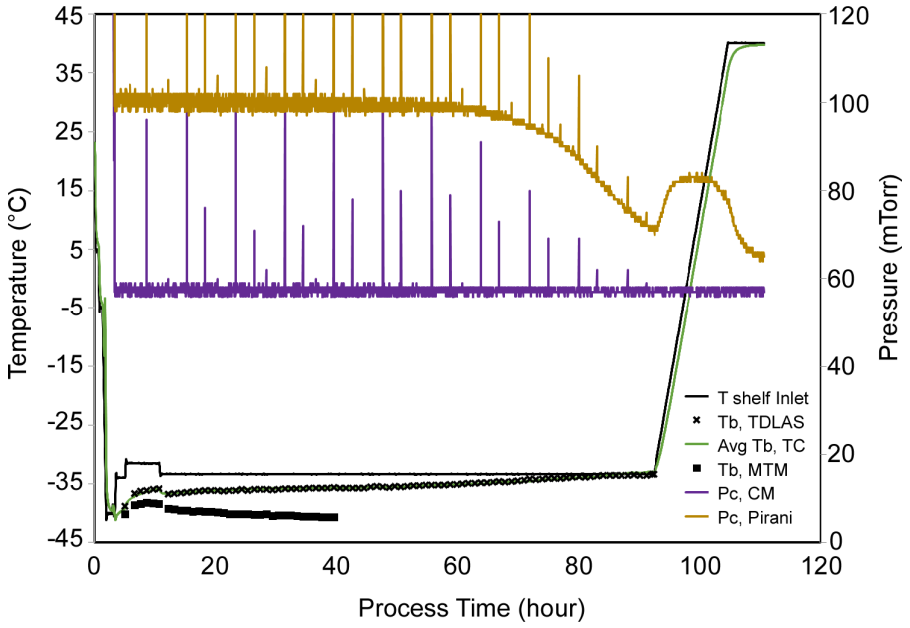

Fig. 3 Lab-scale SMART TDLAS-based Freeze- Dryer cycle developed for 20% w/w sucrose. A single shelf of 160 glass tubing vials (Amcor) filled with 3mL of solution was dried.

For a sucrose concentration as high as 20%, it is expected that significant water reabsorption will occur in the dry product layer during the valve closure and pressure rise event for MTM-based product temperature determinations. This is evident in the product temperatures determined by MTM during the cycle where MTM under-predicts the product temperature compared to thermocouple measurements, with the deviation increasing throughout the

process as the dry layer thickness increases. MTM measurements end 2/3 through primary drying due to low sublimation rates causing insufficient pressure rise for MTM measurements later in the cycle. However, TDLAS-determined product temperatures are accurate throughout the entire primary drying process. Pressure spikes in Fig. 3 and temperature spikes in Figs. 3 through 6 are a result of the isolation valve closures during MTM measurements.

Experiments were conducted in both lab-scale (LyoStar 3) and pilot-scale (LyoConstellation S20) freeze-dryers to demonstrate the application of SMART TDLAS-based Freeze-Dryer technology on multiple scales. Fig. 4. shows primary drying cycles determined by the SMART TDLAS-based Freeze-Dryer for 5% sucrose formulations and 20% BSA:Sucrose at a 1:1 ratio formulations in the lab-scale freeze-dryer. During sucrose drying, 112 vials were filled with 3 mL of solution, and a ring of empty "dummy" vials surrounded the product vials to lessen the radiation input from the warm walls in an attempt to have all vials behave as center vials. The K_v value input to the SMART TDLAS-based Freeze Dryer cycle development algorithm reflected a batch average value determined using the same vial fill configuration. During drying of the 160 vials filled with the BSA:sucrose formulation, all vials were filled with product and a location dependent weighted average value of K_v was used during the SMART TDLAS-based cycle development.

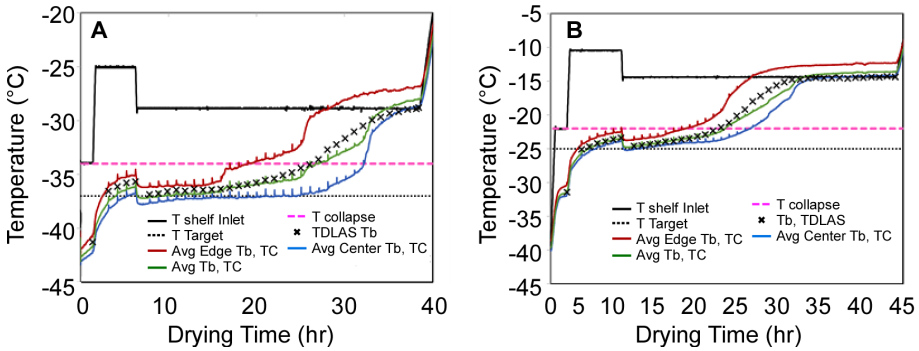

Fig. 4 Lab-scale primary drying recipes determined by the SMART TDLAS-based Freeze-Dryer for a) 5% w/w sucrose and b) 20% w/w BSA:Sucrose at a 1:1 ratio. For 5% sucrose, a single shelf of 112 glass tubing vials (Schott) filled with 3mL of solution was dried. For 20% BSA:Sucrose, a single shelf of 160 glass tubing vials (Schott) filled with 5mL of solution was dried.

Fig. 5 shows primary drying cycles for the same formulations in the pilot-scale freeze-dryer, without the use of "dummy" vials. The variation in number of vials was due to the vial configuration on the shelf, and for each experiment the shelf was fully loaded with the chosen configuration. K_v was determined experimentally prior to each cycle using an identical configuration. For both the low and high solids content formulations at both scales, the TDLAS sensor accurately determined product temperatures as compared to thermocouple probes (within ±1°C for the first 2/3 of primary drying when all vials are undergoing drying). In all cases, the shelf temperature set points calculated by the SMART TDLAS-based freeze-dryer maintained the average product temperature within ±2°C of the target product temperature and below the collapse temperature. Critically, the edge vial temperatures, the warmest vials due to radiative heat inputs from the non-temperature-controlled walls, were maintained below the collapse temperature while undergoing drying. The rise in product

temperature beginning approximately midway through primary drying is due to edge vials completing drying and warming to at or above the shelf temperature. Temperature readings above the shelf temperature are due to wall radiation heat input to these vials. Once the ice is removed, the glass transition temperature of the dried product is raised. Therefore, these product temperature increases are not detrimental to the product structure.

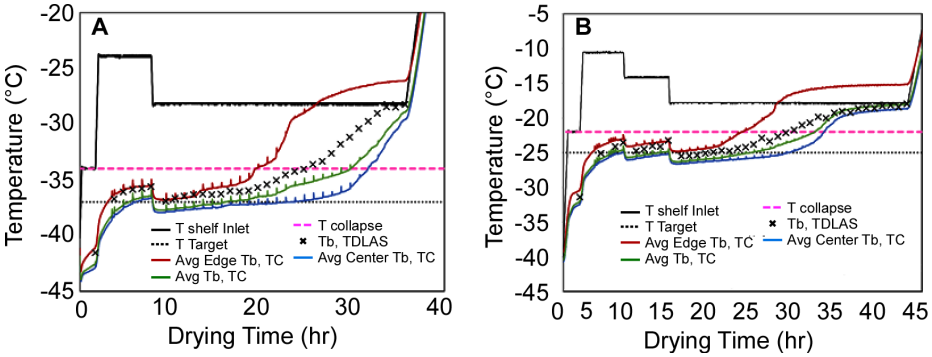

Fig. 5 Pilot-scale primary drying recipes determined by the SMART TDLAS-based Freeze-Dryer for a) 5% w/w sucrose and b) 20% w/w BSA:Sucrose, 1:1. For 5% sucrose, a single shelf of 382 glass tubing vials (Schott) filled with 3mL of solution was dried. For 20% BSA:Sucrose, a single shelf of 480 glass tubing vials (Schott) filled with 5mL of solution was dried.

Experiments were conducted to determine the reproducibility of the cycles developed by the SMART TDLAS-based freeze-dryer. Two experiments were conducted for 5% sucrose (Fig. 6a) and 5% mannitol (Fig. 6b) in the lab-scale freeze-dryer. As shown in Fig. 6, shelf temperature set points and product temperatures were consistent between the two cycles for both formulations.

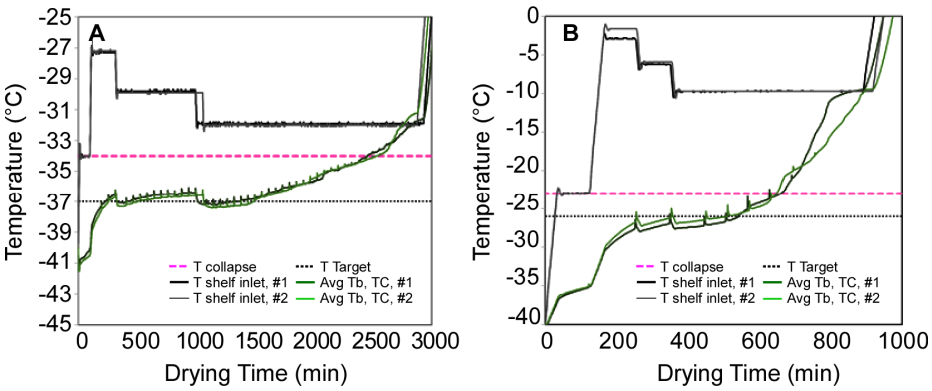

Fig. 6 Repeat lab-scale primary drying recipes determined by the SMART TDLAS-based Freeze-Dryer for a) 5% sucrose and b) 5% mannitol, a single shelf of 112 glass tubing vials (Amcor) filled with 3mL of solution was dried.

4. Conclusions

Replacement of the MTM product temperature measurement data with the TDLAS-based data enables application of the SMART TDLAS-based Freeze-Dryer technology to highly concentrated, amorphous, biopharmaceutical formulations not successfully handled using the MTM technique due to water reabsorption during the pressure rise measurements. The TDLAS measurement technology enables accurate determination of the product temperature for all formulations tested, providing a measurement accuracy of approximately +/-1°C for nearly all formulations throughout the first 2/3 of the primary drying phase of lyophilization when all vials are undergoing drying. This measurement accuracy enables the SMART TDLAS-based Freeze-Dryer to automatically develop reproducible primary freeze-drying cycles for emerging biological products using a single experiment, independent of amorphous or crystalline structure and solid weight percent in the formulation. This is a critical enabling technology for use by process engineers and scientists with limited freeze-drying experience. Because the TDLAS-based measurement technique and the SMART TDLAS-based Freeze-Dryer technology are applicable for all scale lyophilizers, it will also enable rapid process scale-up from laboratory to pilot scale freeze-drying demonstrations.

5. Acknowledgements

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6. References

[1] Tang, X.C.; Pikal, M.J. Design of freeze-drying processes for pharmaceuticals: practical advice. Pharmaceutical research 2004, 21 (2), 191-200.

[2] Pikal, M.J. Freeze-drying of proteins. Part I: process design. BioPharm 1990, 3 (8), 18-27.

[3] Tang, X.C.; Nail, S.L.; Pikal, M.J. Freeze-drying process design by manometric temperature measurement: design of a smart freeze-dryer. Pharmaceutical research 2005, 22 (4), 685-700.

[4] Schneid, S.C.; Johnson, R.E.; Lewis, M.L.; Stärtzel, P.; Gieseler, H. Application of process analytical technology for monitoring freeze-drying of an amorphous protein formulation: use of complementary tools for real-time product temperature measurements and endpoint detection. Journal of pharmaceutical sciences 2015, 104 (5), 1741-1749.

[5] Gieseler, H.; Kessler, W.J.; Finson, M.; Davis, S.J.; Mulhall, P.A.; Bons, V.; Debo, D.J.; Pikal, M.J. Evaluation of tunable diode laser absorption spectroscopy for in-process water vapor mass flux measurements during freeze drying. Journal of Pharmaceutical Sciences 2007, 96 (7), 1776-1793.

[6] Costantino, H.R.. Excipients for Use in Lyophilized Pharmaceutical Peptide, Protein. Lyophilization of biopharmaceuticals 2004, 2, 139.

[7] Liao, X.; Krishnamurthy, R.; Suryanarayanan, R.. Influence of processing conditions on the physical state of mannitol—implications in freeze-drying. Pharmaceutical research 2007, 24 (2), 370-376.