




Best Practices and Guidelines (2022) for Scale-up and Technology Transfer in Freeze Drying Based on Case Studies. Part 2: Past Practices, Current Best Practices, and Recommendations

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Abstract

Scale-up and transfer of lyophilization processes remain very challenging tasks considering the technical challenges and the high cost of the process itself. The challenges in scale-up and transfer were discussed in the first part of this paper and include vial breakage during freezing at commercial scale, cake resistance differences between scales, impact of differences in refrigeration capacities, and geometry on the performance of dryers. The second part of this work discusses successful and unsuccessful practices in scale-up and transfer based on the experience of the authors. Regulatory aspects of scale-up and transfer of lyophilization processes were also outlined including a topic on the equivalency of dryers. Based on an analysis of challenges and a summary of best practices, recommendations on scale-up and transfer of lyophilization processes are given including projections on future directions in this area of the freeze drying field. Recommendations on the choice of residual vacuum in the vials were also provided for a wide range of vial capacities.

Keywords freeze drying · lyophilization · scale-up · technology transfer

Introduction

Developing efficient and robust lyophilization processes at commercial scale remains a nontrivial task even in the twenty-first century. While many tools and approaches were developed to assist in lyophilization cycle optimization at laboratory scale [1], establishing commercial processes at any scale is a complicated process due to numerous

challenges that could arise with the change in scale. The challenges during lyophilization process scale-up and transfer were outlined and discussed in the first part of this paper [2]. It was demonstrated that due to differences in refrigeration capacity, shelf mass, and heat transfer coefficients between commercial and laboratory dryers, the same process parameters in laboratory scale may not be applied to commercial dryers. Some phenomena such as vial breakage

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or vial fogging, not seen in laboratory experiments, could suddenly appear at the commercial scale. The purpose of the second part of this paper is to share past (good and bad) practices and provide some guidance for the industry based on the experience of contributors. It was shown that transferring the same process set points from dryer to dryer (“as is”) may not be a great idea especially for collapse sensitive products. Modeling of the lyophilization process was shown to be a good practice given multiple examples of successful scale-up and transfer. The importance of regulatory considerations during scale-up and transfer was discussed along with the guidance on demonstration of equivalency between dryers. The long-time discussion in the freeze drying community on the proper choice of residual vacuum in the vials after stoppering (or as it is often called “head space pressure”) was addressed with the data calculated for different sizes of vials and fill volumes. The principles of process scale-up/transfer were summarized followed by detailed recommendations based on co-authors’ experience.

Past Practices

Unsuccessful/Non-rational/Trial-and-Error-Based Approaches (Cycle “Transfer As Is”)

In the early days of lyophilization process development (per author’s experience up to end of the 1980s), for both laboratory and commercial scales, trial and error-based approaches were common. Reliable PAT tools (i.e., comparative pressure measurement, pressure rise test) were not available. With advances in process understanding and development

of the Smart™ Freeze Drying software [3] and implementation of reliable fast responding capacitance manometers and isolation valves, the design of optimized lyophilization cycle became routine. However, those tools were not available for the large-scale dryers due to technical limitations. Process transfer and scale-up may still resemble a black box without the use of in-process data and thorough characterization of the lyophilizers using minimum controllable pressure and understanding the heat transfer coefficient (K_v) for the specific vials.

Figure 1 shows a couple of examples of inefficient transfer and scale-up. When a relatively aggressive cycle was transferred from one commercial dryer to another, pressure control was lost at the peak of sublimation (Fig. 1, left panel) when the same cycle parameters were used. Later analysis showed that the second dryer has a smaller and longer duct between chamber and condenser than the first dryer, a factor that was not taken into account during the transfer process. Another example is shown in Fig. 1 (right panel), where the cycle was transferred from a laboratory to a commercial dryer using the same process parameter set points (so-called transfer as is) using the same container closure. In this case, primary drying on the pilot dryer was not complete prior to the ramp to the secondary drying resulting in product collapse due to lower vial/shelf heat transfer coefficient on this dryer.

Another example of failure using an “as is” approach is shown in Fig. 2.

The cycle shown in Fig. 2 was scaled up using an “as is” approach; that is, the same shelf temperature/chamber pressure/time profile was used at both laboratory and manufacturing scale. The appearance of the product from both

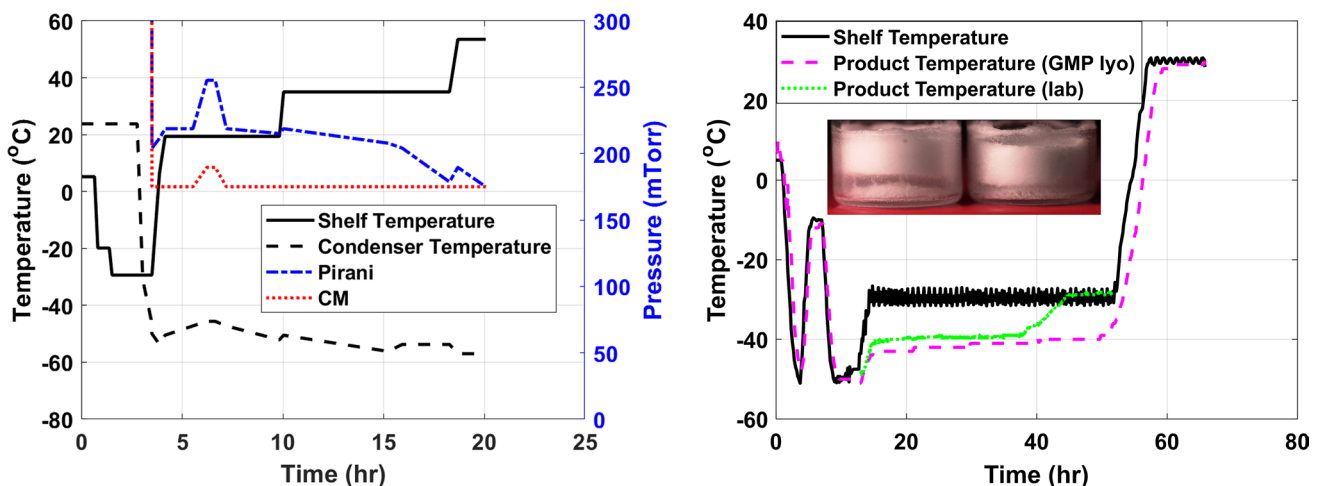


Fig. 1 Left panel represents the case with pressure control loss (seen as a hump in the pressure readings) when the cycle was transferred from one commercial dryer to another. Right panel represents another case where differences in product temperature profiles during cycle transfer from the laboratory (magenta

dashed line) to the pilot dryer (green dotted line). The inset picture of vials with collapsed product is shown. Both cases are examples of cycle transfer, referred to “as is,” where the same process parameters were used without accounting for the differences between dryers

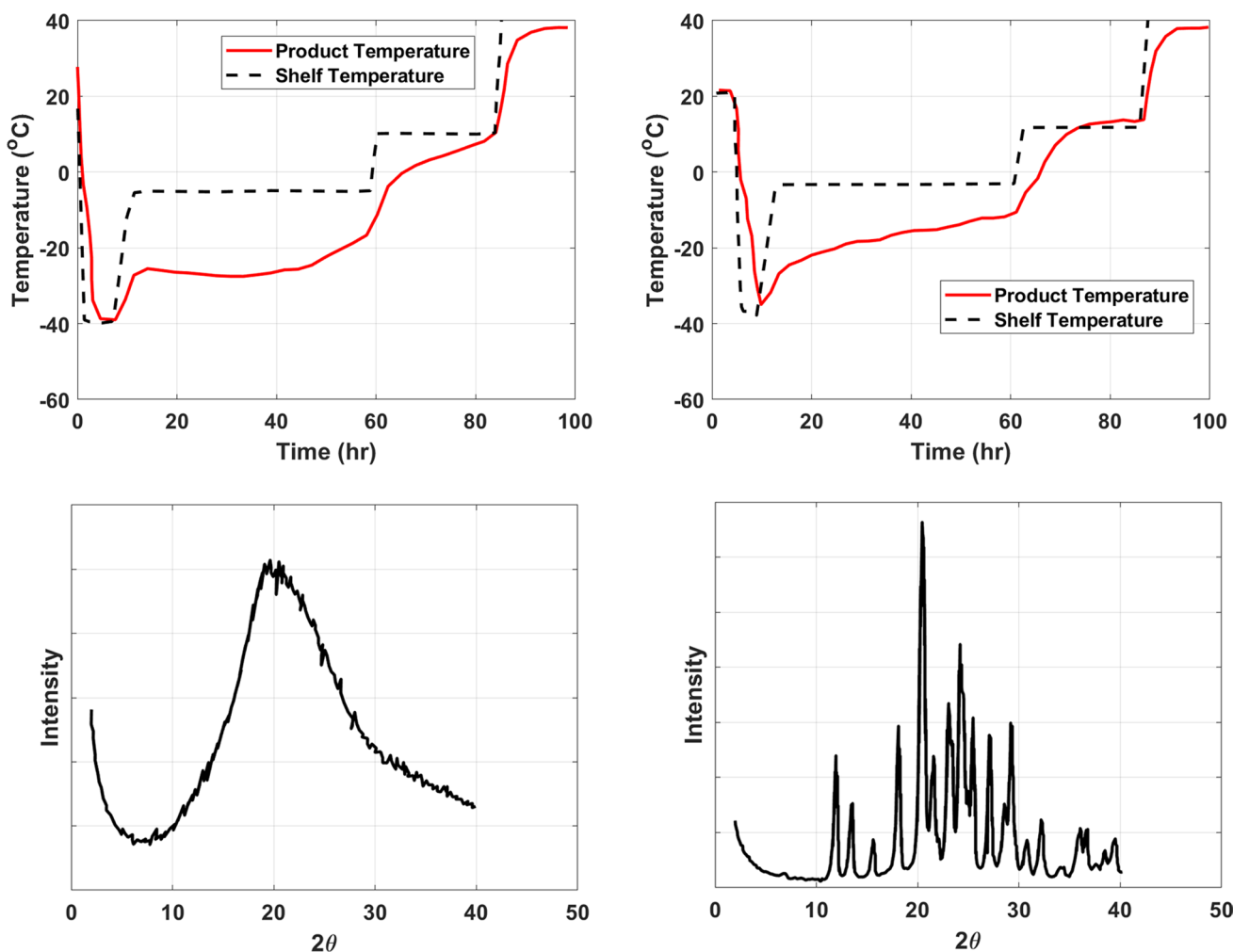


Fig. 2 Lyophilization cycle traces of proprietary small molecule drug (left top panel—laboratory cycle, right top panel—large-scale cycle). Bottom left panel represents X-ray powder diffraction pat-

tern of lyophilized material after laboratory-scale experiment while right bottom panel shows XRPD traces of the same material after the commercial cycle

laboratory and manufacturing scale batches was the same—a white to off-white powder with uniform color and texture, with no sign of cake shrinkage or collapse—both were considered pharmaceutically acceptable. The difference between the two scales became apparent when reconstitution time was measured—the reconstitution time of the product from full-scale manufacture was about three times the reconstitution time of the laboratory-scale batches. Further analysis by X-ray powder diffraction revealed a striking difference, as shown in Fig. 2. The laboratory batch (left panel) is amorphous, whereas the scaled-up batch (right panel) shows well-defined crystallinity.

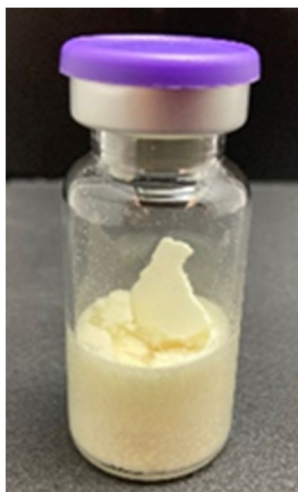
This is an unusual case for at least two reasons. First, the crystalline form of an API is usually preferred because the crystalline state is usually more chemically stable than the same drug in the amorphous state. There appears to be no chemical stability issue with this drug as an amorphous

solid. However, the crystalline form dissolves more slowly than the amorphous form. In this case, the amorphous form is preferred. Second, freeze dry microscopic analysis (data not shown) revealed that the drug crystallizes rapidly from collapsed, or partially collapsed, freeze-dried solid, but not from a freeze-dried solid that has retained the microstructure established by freezing.

It appears that primary drying was not complete in the commercial-scale (Fig. 2—right top panel) cycle when the secondary drying process conditions were initiated. This likely triggered the onset of collapse, with relatively rapid crystallization of the API which resulted in the undesired product attribute of an extended reconstitution time.

Another case where the “as is” approach failed was scaling up from partial load to full load during manufacturing. The cycle transferred well from the development laboratory to partial dryer but flaking or slicing off the top layer of the

Fig. 3 Flaking of solid during commercial batch



solid was observed when the product was scaled up to a full dryer using all shelves (Fig. 3). Insufficient primary drying time was found to be the root cause of this failure.

Authors also witnessed cases when random, not science-based choice of process parameters was used during scale-up and transfer. The cost of failure can be weeks or even months of commercial manufacturing time. One of the authors reports the case when at least 20 cycles at the commercial scale were performed to ensure a robust commercial process.

Change of Container Closure Without Adjusting Cycle Parameters (“As Is”)

Another example involves a change in vial for a lyophilized product. The registered vial was discontinued by the vendor and replaced with a similar 50-mL vial from same vendor located at another site. A commercial-scale product run was performed using the “as is” cycle with same lyo cycle

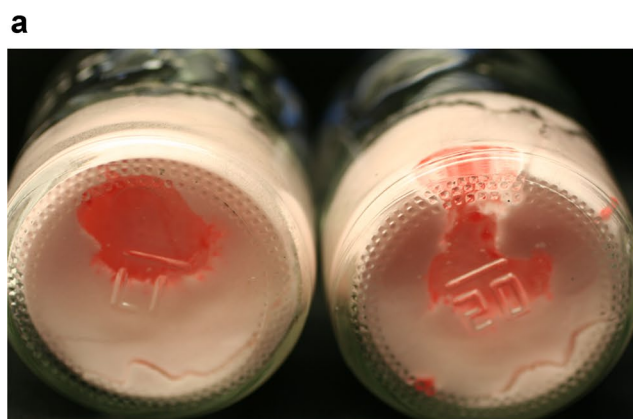


Fig. 4 Appearance of meltback in the lyophilized product using as is lyo recipe with new vials (left panel, **a**). Cycle traces (right panel, **b**) demonstrate longer primary drying with new vials. This was due to

Table I Vial Heat Transfer Coefficient of Discontinued (Old) and Proposed (New) Vial at 150 mT

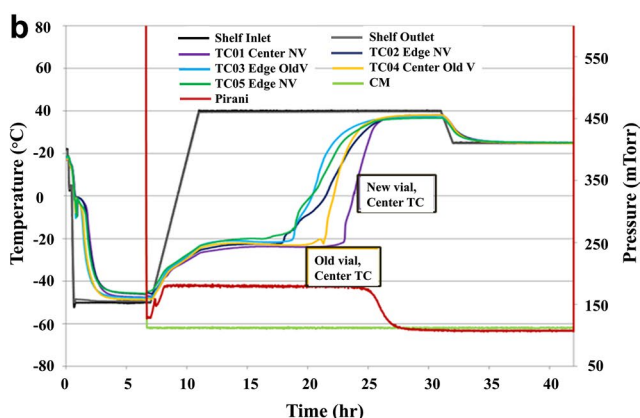
Glass vial	Center vial K_v (cal/ $K*s*cm^2$)	Edge vial K_v (cal/ $K*s*cm^2$)
Old discontinued	3.17E-04	4.01E-04
New proposed	2.48E-04	3.30E-04

parameters with the new vial. Primary drying was not complete before the cycle was stopped as shown by meltback in some vials upon inspection (Fig. 4a). Further investigation indicated that primary drying was longer based on the product temperature data (Fig. 4b). This was because the new vial had a lower K_v (Table I) meaning that hold times needed to be longer if using the same cycle parameters (shelf temperature and chamber pressure) or the critical process parameters needed to be adjusted to keep the same cycle time with no product meltback.

In another example of the impact of changing vials of the same size and vendor, but a different manufacturing location, no fogging was observed during the lab-scale development and PQ (process qualification) batches with the old vial, while fogging was observed with the new vial (Fig. 5). A specification of no fogging for the drug product meant that the batches had to be rejected.

Successful Practices

Conservative approaches to lyophilization are common in the pharmaceutical industry due to the significant cost of manufacture and expensive products that need to be lyophilized (especially biologics). Typically, the same process set points are used in process scale-up; however, all steps (freezing hold, primary and secondary drying steps)



lower vial heat transfer coefficient (Table I) of new vials. Abbreviations: TC—thermocouple, CM—capacitance manometer, NV—new vial, old V—old vial

Fig. 5 Fogging observed in new vials



were longer than in laboratory dryers due to increase in thermal load at full scale. As it is commonly assumed, the reduction of particulates during manufacturing (typical for GMP) leads to a higher degree of supercooling as compared to a laboratory-scale unit, resulting in an increase in cake resistance causing an increase in product temperature during primary drying. In one example, almost 3 deg higher product temperature was measured at GMP clinical scale. The temperature of supercooling (T_{sc}) was measured to be -19.9°C at clinical scale (Tempris wireless sensors were used—data not shown), whereas the T_{sc} was found to be -16.4°C at laboratory scale (T-type thermocouples were used) for the same formulation, fill volume, protein concentration, and same vial using the same cycle set points. The presence of the temperature sensors could itself be a center of nucleation and, therefore, the true difference in degree of supercooling between laboratory and GMP products may be greater. A higher product temperature (T_p) during primary drying and an increase in primary drying time was confirmed for the clinical scale batch, as shown in Fig. 6.

The product contained sucrose and the higher degree of supercooling likely resulted in an increase in product resistance during primary drying. Fortunately, the set points for the cycles were conservative and likely mitigated any risk to cake appearance (some shrinkage was observed as expected for sucrose cakes).

The criterion in this case was the comparability of laboratory and commercial materials regarding product critical attributes (moisture, high molecular weight, reconstitution time, etc.) which was achieved in this particular case and could be achieved in most cases when formulation is robust, and the cycle is conservative.

While this particular case shows an example of successful transfer “as is,” the opposite examples shown in Figs. 1, 2, 3, 4 and 5 demonstrated that failure is more likely. For example, more shrinking or possibly collapse could be expected when using more aggressive cycles during scale-up and transfer.

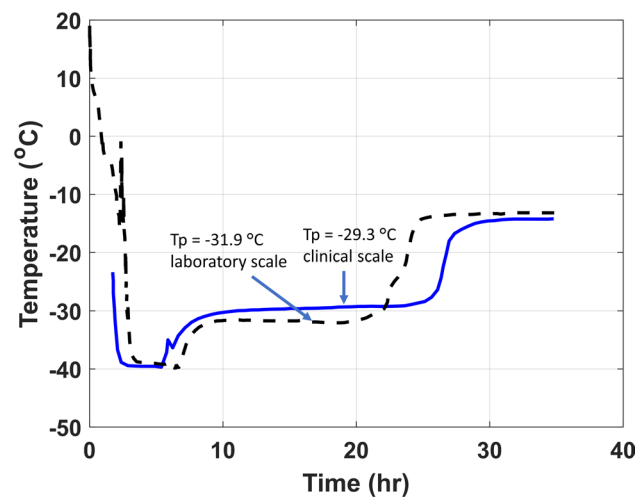


Fig. 6 Comparison of product temperature during primary drying for a lyophilized placebo product manufactured at laboratory scale and at clinical scale at same cycle set points with product temperature measured in the similar vial position (center of shelf)

Current Approaches

Leveraging Historical Experience (Empirical and Risk-Based)

Leveraging historical experience with lyophilization of similar products in the same dryer is somewhat of a continuation of the past successful practices. If product characteristics and container closures are similar and the freeze dryer load is the same or smaller, the same cycle can be used. The manufacturing of clinical batches of two different drug products containing the same platform formulation is an example that used this approach. The thought process of “change as little as possible” therefore provides an easier path to scale up (in the short term) but it has limitations. The authors recommend leveraging the data to perform risk assessment and provide recommendations for the process transfer of the same product under identical conditions, keeping in mind that an engineering batch may still be required for confirmation.

Implementation of PAT Tools at Full Scale: Improving Product Appearance Through the Use of a Pirani Gauge as Best Practice for Determination of the End of Primary Drying

In one of many successful examples, a formulation containing sucrose, tris, and an active ingredient filled with 1 mL into 3-mL vials was transferred to a third-party manufacturer. There were two challenges resulting from this. The first was that the choice of formulation components resulted in a critical product temperature of -38°C (measured by freeze drying microscopy). The low critical product temperature is difficult to

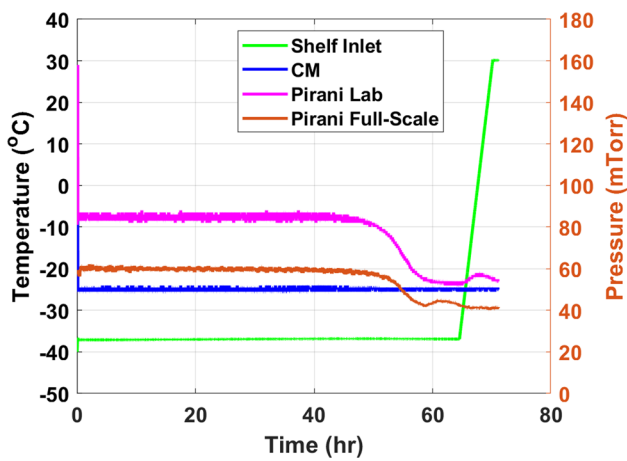
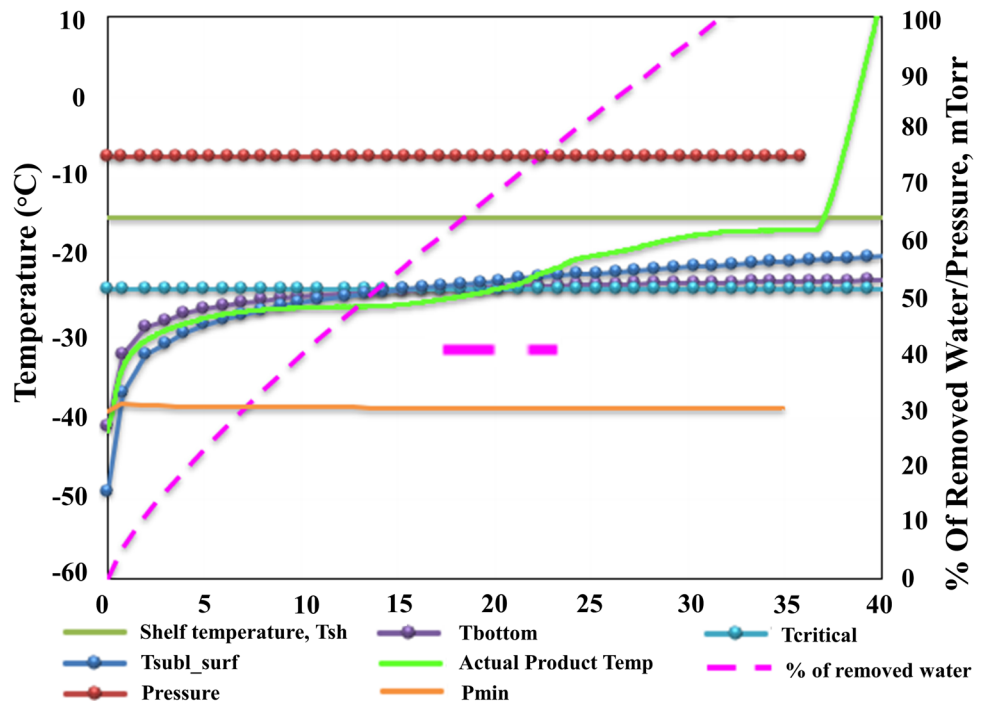


Fig. 7 In-process data for the primary drying cycle from a laboratory-scale lyophilizer (Pirani lab) compared with a full-scale lyophilizer (Pirani full scale) showing that both dryers required approximately 60 h for completion of primary drying

maintain at full scale and leads to a long primary drying cycle to prevent collapse. The next challenge was that the product was transferred to a lyophilizer that was not equipped with a Pirani gauge for comparison with a capacitance manometer. The lack of a Pirani gauge meant that the primary drying cycle had to be based on time rather than relying on in-process data to determine the end point of primary drying. The total cycle time provided by the client for full-scale lyophilization was 70 h with 46 h designated for primary drying conducted using a shelf temperature of -37°C with chamber pressure of 40 mTorr.

Fig. 8 Laboratory data output demonstrating accuracy of product temperature prediction using steady-state primary drying model



Most of the product removed at the end of the cycle exhibited poor appearance with severe shrinking. Laboratory-scale data demonstrated that the Pirani gauge pressure starts to decrease after approximately 50 h of primary drying time with primary drying complete after 60 h. A Pirani gauge was installed on the full-scale lyophilizer, and the process was advanced after evidence of the Pirani gauge pressure becoming similar to the pressure of the capacitance manometer. The data were compared with the laboratory-scale data and the times required for completion of primary drying were similar (Fig. 7).

The case study demonstrates the best practice of using the differences in pressure between the capacitance manometer and Pirani gauge to identify the end point of primary drying.

Utilization of Modeling for the Primary Drying Step

Even relatively simple steady-state models of lyophilization processes [4] could significantly reduce risk during process scale-up and transfer.

Models for primary drying can closely predict how the product temperature will track during primary drying. The models are created using parameters such as the K_v for the vial and the P_{min} for the dryer that are experimentally determined at laboratory scale. The data are entered into the model for commercial scale to confidently predict parameters that will ensure the product temperature is well below the point of failure. This avoids the need for several costly and time-consuming engineering runs at commercial scale.

Figure 8 demonstrates how the prediction works and shows data generated at laboratory scale (for a specific vial

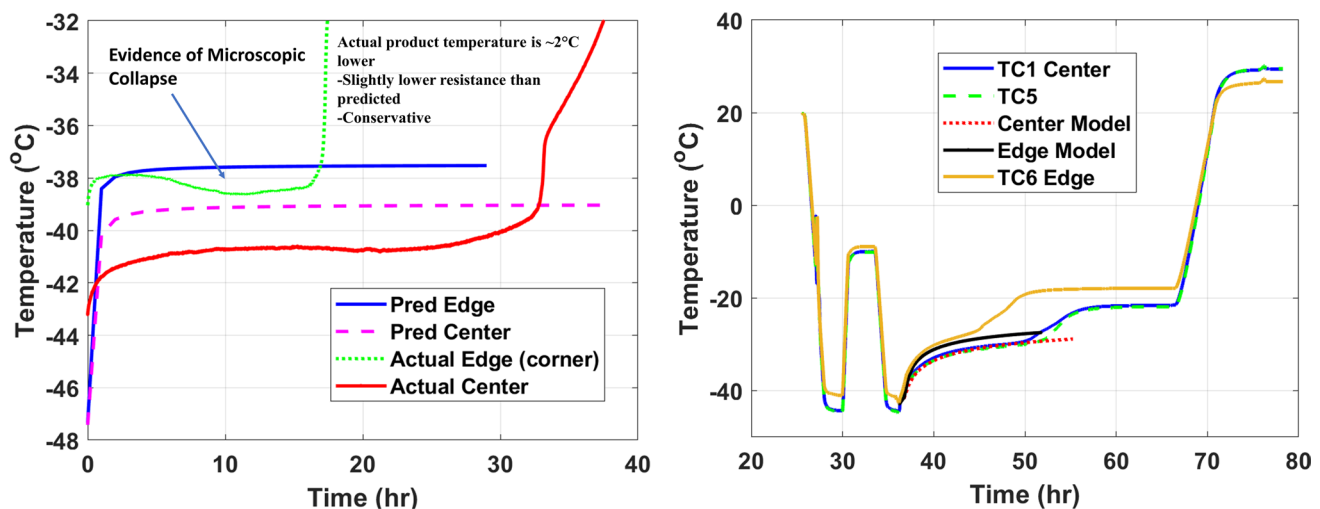


Fig. 9 Utilization of modeling for process transfer from one site to another of sucrose-based drug product containing a low amount of active ingredient (<1%). Process parameters were adjusted accounting for the differences in the dryers (left panel). The right panel repre-

sents scale-up drug product with medium Tc (about -26°C) and high solid content (14%). In both cases, vial heat transfer coefficients and minimum controllable pressure were experimentally determined for the dryers

type, with known K_v) with a dryer with experimentally determined P_{\min} , demonstrating verification of the model with an overlay of predicted *versus* actual product temperature.

Figure 9 shows an example of cycle scale-up of sucrose-based formulations (left panel—low collapse temperature/low solid content, right panel—medium collapse temperature/high solid content) when process transfer was performed with the assistance of a primary drying model followed by a successful single engineering run. The engineering run is a trial or test run of the lyophilization process carried out for process optimization and to ensure the final product meets the desired specifications. It is typically performed on the clinical or commercial freeze dryer, intended to manufacture this particular product, and may include testing of various process parameters (in particular product temperature) to refine and optimize the lyophilization process prior to PV campaign. The freeze dryer was characterized prior to the engineering run by determination of the minimum controllable pressure and the K_v for the vial. Prior to engineering run, modeling of product temperature profiles was performed accounting for differences in cake resistance and vial heat transfer coefficient. In both cases (shown in left and right panels in Fig. 9), the difference in calculated product temperature and actual product temperature did not exceed 2°C for both edge and center locations. Based on successful engineering runs for both products, shown in Fig. 9, no adjustments of cycle parameters were made for following PV campaigns which produced both products with acceptable quality.

An earlier example of utilization of primary drying modeling for scale-up was presented by Kramer *et al.* [5]. In that study, a semi-empirical computational 2-dimensional heat and

mass transfer model originally developed by Mascarenhas *et al.* [6] and commercialized as a Passage software was used to calculate the product temperature and duration of the primary drying for laboratory and pilot freeze dryers. Primary drying conditions were established at laboratory scale using Smart FD, and then the computer model was used to select shelf temperature for the pilot freeze dryer, in order to match the product temperature profiles on the laboratory and pilot scales. The calculated pilot shelf temperature was 10°C higher than that for the laboratory scale (26.2°C vs. 16°C , respectively, calculated for the edge vials in both scales) due to at least 12% (2.85×10^{-4} pilot/ 3.24×10^{-4} laboratory) lower vial heat transfer coefficient. The recommended primary drying conditions for the pilot scale were confirmed experimentally, with a good agreement observed between the modeling results and the experimentally determined product temperature and primary drying time values. The same Passage model was utilized to establish design space for a different product on a commercial scale [7].

Modeling proved to be a very useful approach that can be used to evaluate the possible impact of process parameters on the product quality (by calculating product temperature profiles) during the commercial process and, especially, during process deviations. It requires, however, extensive experimentation to generate input parameters into the model. At minimum, it should include determination of the following:

- Minimum controllable pressure (MCP)/choked flow conditions. This will allow assessment if pressure control could be lost (example is shown in Fig. 1, left panel) when the cycle is transferred to the commercial dryer. One could make a decision on either reduction of batch size

- (less probable) or modification of ramps, pressure/shelf temperature set points to ensure stable pressure control.
- b) Maximum sublimation rate (MSR) (max sublimation rate). This will allow assessment if the dryer could be overloaded (condenser temperature exceeding -40°C , for example for the oil-sealed old vacuum pumps) or if the refrigeration system could fail when an aggressive cycle is used at commercial scale.
 - c) Shelf surface temperature distribution as a function of load. This will allow estimation of the time needed for equilibration at the freezing set point to ensure homogeneity of the product temperatures. In this case, flat self-adhesion thermocouples could be attached to the bottom of shelves at different locations. Thermocouples could also be placed in the vials, located on the edge and center of the shelves. To minimize complexity of thermocouple placement, in case of automated loading, wireless sensors could be used.
 - d) Vial (container) heat transfer coefficient (K_v or $K_{v,s}$) as a function of pressure and position (edge effect). Ongoing efforts on best practice recommendations for equipment qualification are summarized in [8].

Execution of items (a-c) could take up to a week while inclusion of heat transfer coefficient measurements could increase the duration of characterization study up to 2 weeks. Even though the characterization of dryers and container closures takes some time, the input parameters to the model do not significantly change with time, assisting with multiple transfers.

There are examples in the literature where minimum controllable pressure could be calculated using computational fluid dynamic modeling [9, 10]. This provides an option of using CFD modeling and extensive OQ data to minimize down time of commercial freeze dryers needed for lyophilization characterization.

Addressing Challenges During Scale-up and Transfer

Stochastic Nature of the Nucleation Process

Ice nucleation is a random process that is heterogeneous under the conditions used for lyophilization. Nuclei often form as a result of a seed present in the solution. The seed can be a dust particle and/or imperfections in the container holding the solution [11–13]. The homogeneous ice nucleation temperature for water is approximately 235 K [14], whereas the ice nucleation temperature for everyday systems is always higher because of the presence of possible nucleation seeds. Solutes included in freeze-dried solutions will decrease the equilibrium melting point and may affect the ice nucleation temperature, for both homogeneous and heterogeneous nucleation mechanisms.

The relationship between equilibrium ice melting point (water liquidus) and heterogeneous nucleation temperature, T_{het} , is described by [15] as

$$\Delta T_{het} = k\Delta T_m \quad (1)$$

where ΔT_{het} is the depression of the temperature of heterogeneous nucleation of hexagonal ice by a solute, ΔT_m is the freezing point depression, and k is an empirical constant. k values were reported between 1.5 and 4.4 [16] and 1.3 to 2.6 [17] depending on solution composition and ice nucleation agents added. Correlation between the equilibrium melting point and homogeneous ice nucleation temperature can be described by a similar equation [18]; although with lower k values between 1 and 2.

Note also that studies of such probabilistic processes as nucleation would require large statistics, with tens and hundreds of replicates [19], as well as a control of the type and concentration of centers of heterogeneous nucleation.

There is no definite agreement on relationships between cooling rate and ice nucleation temperature. While it has been suggested that slow cooling causes larger supercooling [20], and also reported that cooling rate (range 0.6–40 K/min) did have some impact on the homogeneous ice nucleation temperature in solutions of LiCl with concentration above 6.8 mol% [21], there are also reports to the contrary. Indeed, no impact of the cooling rate on the homogeneous ice nucleation temperature was observed for LiCl solutions at 5 mol% [21]. Furthermore, it was reported that an increase in the cooling rate from 0.1 to 1000 K/min resulted in only 2°C difference in the heterogeneous ice nucleation temperature and 4°C in homogeneous ice nucleation [22]. No trend in the ice nucleation temperature was observed with cooling rates of 0.5– $3.2^{\circ}\text{C}/\text{min}$ [23], 0.07 – $7^{\circ}\text{C}/\text{min}$ [24], and 0.05 – $1^{\circ}\text{C}/\text{min}$ [25]. Also, nucleation rate coefficients of ice on kaolinite did not demonstrate any significant difference for cooling rate 0.8–10 K/min [26]. There are anecdotal reports of studies in which impact of cooling rate on ice nucleation temperature was indeed observed; unfortunately, results of these studies have not yet been published.

Since there is no solid evidence that cooling rate can be used to govern ice nucleation temperature, annealing [27] or controlled nucleation technologies [28–30] may be utilized to reduce a negative impact of heterogeneity of product properties (specific surface area, etc.) created by stochastic nature of nucleation process.

Cake Resistance Differences Between Products Made in Laboratory Versus Products Made in Clean Class A Commercial Manufacturing Conditions

Differences in the particulate matter levels of the environment could impact a lyophilization cycle during the freezing step when ice crystals are formed. For example, the grade A and grade B environments contain substantially

fewer particles than an unclassified laboratory environment. A reduction of particles that could serve as seeds for ice nucleation results in higher degrees of supercooling in the manufacturing area than in the laboratory. High degrees of supercooling leave less time for growth of ice crystals. Sublimation of smaller ice crystals creates smaller pores in the drying solid, increasing the resistance to mass transfer of water vapor during primary drying. An increase in resistance to the mass transfer of water vapor can increase drying time, but can also increase product temperature. Failure of the drying process can occur if the increase in product temperature approaches or exceeds the critical product temperature. Ideally, one would prefer to use similar conditions in both laboratory and commercial scales but maintaining grade A conditions in the laboratory is impractical and expensive. One way to mitigate the differences in ice crystal size in manufacturing is by annealing, post-freezing [27]. The product is held at a temperature above T_g' during annealing to provide additional time for crystals to grow in size (Ostwald's ripening). This approach is relatively simple and requires no modification to the equipment. The second approach is to perform controlled ice nucleation by either the rapid depressurization method [28] or ice fog method [30] both during development in a laboratory scale and during scale-up in production. Introduction of a controlled ice nucleation step, however, requires special fitting to the equipment which may not be financially viable and may require equipment requalification and resubmission of regulatory documents. Alternatively, vacuum-induced nucleation method [29] could potentially be implemented at commercial scale but it might require software adaptation as well as a demonstration at a large scale.

A small caveat to keep in mind with the inclusion of an annealing step, is, although primary drying time is optimized, the smaller specific surface area left behind after sublimation, results in a requirement for a longer secondary drying step which was observed by some of co-authors [1].

An alternative option to mitigate the effect of increase in product temperature during primary drying as a result of the GMP factor is to either decrease the shelf temperature or chamber pressure set point during the primary drying step to avoid the product exceeding its critical product temperature. Indeed, a higher degree of supercooling in GMP environment results in smaller ice crystals in the frozen product, which leads to smaller pores and increased product resistance to water vapor mass transfer during the primary drying process. Consequently, given the same process conditions, shelf temperature, and chamber pressure, the resultant product temperature will be higher, which may result in product failure. Either of these or both can be decreased in order to maintain the product temperature below the critical value.

Vial Breakage

Glass vials for parenteral packaging differ by composition, manufacturing method, and the degree of thermal expansion. Three types are available that include type I borosilicate glass, type II soda lime glass treated with ammonium sulfate to remove metal ions, and type III untreated soda lime glass. These are divided into molded and tubing glass vials. Molded glass vials are manufactured by placing molten glass into a mold, using air to expand the glass into the mold, and annealing the glass to prevent shattering. Tubing glass vials are manufactured from tubing cane that is heated at the ends to form the neck and heel of the vial. Type I glass is also available with 2 different levels for the coefficient of thermal expansion. The vials are identified as 33 coefficient of glass expansion ($32.5 \times 10^{-7}/^{\circ}\text{C}$) and 51 coefficient of glass expansion ($51.0 \times 10^{-7}/^{\circ}\text{C}$). Glass vials with lower numbers of thermal expansion coefficient are more dimensionally stable against thermal expansion stress that can result in cracking. Therefore, 33 expansion glass is a preferable vial material for lyophilized products [31, 32].

Most glass vials must be rinsed, depyrogenated, cooled on a rotating table, conveyed along the filling line, and then often transferred as a pack or shelf load to the lyophilizer. The steps offer many opportunities for vials to contact each other which may result in frictive damage. Frictive damage can result in breakage of glass on the line or weaken glass making it susceptible to breaking during lyophilization [33, 34].

Two factors that contribute to glass breakage during freezing and drying can be addressed during formulation development. The *first* is to suppress crystallization of crystallizable excipient, by having a sufficient amount of amorphous solutes; the ratio of crystalline to amorphous component to avoid the crystallization depends on both particular system and experimental approach [35] and varies between 0.06 and 2. This formulation approach will not work for crystalline bulking agent, because its crystallization is required. The *second* is ensuring that the fill volume is no more than 1/3 of the overfill volume for the vial (as an example, 10R vial can hold 15 mL when filled up to the very top. Therefore, 1/3 of the overfill is 5 mL). High concentrations of crystallizing component along with high fill volumes increase the risk for vial breakage [36–39].

During lyophilization, the design of the freezing and drying cycle is important to prevent glass breakage. Scientific literature suggests that fast cooling rates and cooling to low temperatures, such as -70°C , increase the risk for glass breakage [37, 38]. During freezing, the sample volume initially increases as the result of water to ice transformation and then vial contents solidify into a frozen plug which contracts on further cooling. Due to the

difference in the thermal expansion coefficient of the ice and the glass itself, any further contraction results in the separation of the solid plug from the walls of the vial. This separation is referred to as the break-loose event that can impact the integrity of the vial. The fully solidified frozen plug likely expands on warming during the shelf temperature increase during early primary drying. Additional expansion can occur during solute + water crystallization during such warming.

During stoppering, stoppers are sealed on vials at the end of the lyophilization process by compressing the shelves using a stoppering ram. The ram pressure and duration can be adjusted to prevent crushing glass or having stoppers stick to the shelf above. Stoppered vials are unloaded and the load proceeds to the capper. Caps are crimped onto the necks of vials using a capping machine. Care must be taken when setting up the pressure used to seal the crimps. Excessive pressure can produce cracks in the necks of the vials (Fig. 10).

Different scenarios of vial breakage are summarized in Table II.

Addressing Differences in Dryer Design Between Laboratory and Commercial Dryers Regarding Mass Flow Resistance

Differences exist in the design of laboratory-scale and commercial-scale freeze dryers. The differences have the potential to affect the flow of water vapor from the product chamber to the condenser. If possible, modeling experiments should be completed to better understand the flow of water vapor. It can be done either through computational fluid dynamic (CFD) modeling [9, 10] or by performing sublimation tests at scale [40]. Modeling of the primary drying process [4, 10], using inputs from sublimation tests using CFD, could provide insights into the flow of water vapor during a lyophilization cycle at commercial scale. For example, if a commercial dryer is not capable of maintaining pressure set points, commercial cycle inputs may be adjusted in

order to keep the product below collapse temperature. The commercial cycle inputs include chamber pressure and shelf temperature which are manipulated to control the product temperature.

Designing a new commercial dryer capable of withstanding aggressive cycles is an attractive option for drying products with high failure points during primary drying. For example, if a dryer can maintain a pressure of 200 mT at a normalized sublimation rate of 1.5 kg/sq.m/hr or higher, it could be used for the most aggressive cycles (for example, 40°C shelf temperature during primary drying or higher). Use of CFD modeling at the initial step of dryer conceptual design could be beneficial. Slight modifications of the duct or a mushroom valve, made after CFD analysis of dryer performance, can increase throughput by almost 15%.

Accounting for Edge Effects in Commercial Dryers

Conventional freeze dryers utilize shelves on which vials or other types of containers are placed. Each shelf is cooled with thermal fluid which is used to control the temperature of the solution in the containers on the shelves. However, vials located on the outer rows of the shelves are exposed to the transfer of heat from the walls and door of the freeze dryer. This is referred to as the edge effect and can result in temperature differences between containers located on the edge of the shelf and those containers located in the center [41].

The product in containers located on the edges of a shelf is often warmer than the product in containers located on the center of the shelf. Therefore, development of the lyophilization cycle should consider the temperature at which the product located on the edges of a shelf may experience. The degree of temperature difference between the edge and center is often a function of shelf temperature. Another factor that exacerbates the product temperature differences is the differences in K_v of the same vial across the shelf. This K_v difference is larger when operating at lower shelf temperatures and smaller when using high shelf temperatures [42]. The authors reported a reduction in the edge effect and

Fig. 10 Examples of vial damage resulting from excessive crimping pressure



Table II Summary of Factors Impacting Vial Breakage and Corrective Actions

Scenario	Potential challenges/causes	Corrective action/risk mitigation
Vial handling and manufacturing operations in commercial production	Damage due to friction as vials contact each other on the manufacturing lines	Externally coated vials can help protect vials from damage during handling and commercial-scale operations
Fill volume	Higher fill volume leads to higher breakage rates	Reduce the fill volume to avoid breakage. Interior coating of the vial or silicization (with careful assessment) can dramatically reduce vial breakage
Formulation composition	Crystallization of excipients (e.g., Mannitol + water, NaCl + water) and forces exerted due to volume expansion	Balance ratio of crystalline vs. amorphous excipients in formulation. Use an annealing step if crystalline excipient is desired
Inadequate development of lyophilization process for a particular formulation	Cooling rate and its subsequent impact on solid state transition could result in vial breakage of frozen crystalline solutions. Break-free event during freezing and thermal expansion during drying can affect the integrity of the vial during the process	Robust lyophilization cycle development and control strategy. Use of strain gauges and thermal mechanical analysis can help study the strain profile and provide a knowledge of the thermal events. Making changes to the lyophilization cycle to reduce the strain can help reduce or eliminate vial breakage
Pre-treatment of glass vials	Tubing glass vials are at greater risk for breakage than molded glass vials	Perform detailed glass fracture analysis, glass thickness, and surface analysis
Stoppering and capping/crimping	Extreme hydraulic pressures during stoppering or uneven loading patterns can cause imbalance of forces resulting in vial breakage	Adjust the time-pressure profile to reduce vial breakage

a homogenized drying across the shelf at high shelf temperatures which led to a significant decrease in K_v for edge vials from 18.7 to 13 W/m²/K. The decrease in K_v for the center vials was much less significant, only changing from 9.8 to 7.9 W/m²/K. Overall, the reported outputs from the experiments suggest that an aggressive cycle design comprising a shelf temperature and/or chamber pressure provides a more uniform drying of a batch. However, one limitation of operating an aggressive cycle is that the product temperature should still be maintained below the critical temperature of the formulation.

In theory, the edge effect could be almost eliminated if walls and doors of commercial dryers could be maintained at or near the product temperature, but the authors are not aware of such technology being used in manufacturing. It is, therefore, a common practice (at least within group of co-authors of this paper) to design a conservative cycle that balances both the edge/corner vials not exceeding the product temperature and at the same time allowing for complete drying of center vials. Manufacturers typically resort to this approach unless they are willing to sacrifice the edge vials.

A small-scale micro-freeze dryer [43] is available for laboratory studies and offers the ability to control the wall temperature to mitigate the edge effect. A small number of vials in the micro-freeze dryer can be precisely controlled by a LyoSIM® (temperature-controlled aluminum blocks), to mimic vial-to-vial contacts of manufacturing scale units and match the K_v of center and edge vials. This technology controls the temperature of product in vials so that all vials on the shelf exhibit product temperatures as if they were located on the center of a shelf. Again, this technology is not implemented in manufacturing dryers.

One option for the complete elimination of the edge effect is to introduce a continuous lyophilization process where every single vial is dried at controlled conditions. A spin freeze drying process [44] or continuous freeze drying process with suspended vials [45] provides a path for elimination of the edge effect, significantly reducing freeze drying process time. These technologies, however, are not yet available at full scale.

Estimation of Edge Effect in Commercial Dryers

In the past, the edge effect in commercial freeze dryers was either measured (paper 1) or estimated from experience. This requires process intervention due to use of temperature sensors. A publication by M. Pikal [46] estimated the edge effect in the commercial dryers using previously measured emissivity of shelves and walls of and temperatures of heating surfaces. The heat transfer coefficient of center vials in a commercial dryer can be estimated from Eq. 2 while the edge effect could be estimated from Eq. 3.

Center vials

$$10^4 K_v(Mfg) - 10^4 K_v(Lab) = A_v(e_{Mfg} - e_{Lab}), \frac{Cal}{cm^2sK} \quad (2)$$

where e_{Mfg} is the emissivity of shelves in commercial dryer (typically ~0.3), and e_{Lab} is the emissivity of shelves in laboratory dryer (typically ~0.65).

For the edge vials

$$\Delta K_{vE} = K_v(edge) - K_v(Center) \quad (3)$$

Measurement of emissivity is a relatively easy task and can be done during a site acceptance test (SAT) or during IQ/OQ since SAT is only to confirm intended functionality and test against acceptance criteria post installation. The wall temperature can also be measured during engineering run to estimate the edge effect. This information can be used as an input in the models to design cycle in such way that product temperature in the edge vials will never exceed the critical temperature. Thus, one should consider measuring wall temperature during the regular commercial process given that modern commercial dryers could be equipped with such measurement systems.

Responding to Non-uniformity of Shelf Temperature

Non-uniformity of shelf temperature is a common problem leading to variable temperature distribution over the entirety of the shelves. Temperature differences across and between shelves lead to differences in product temperature between

the vials. The differences in product temperature create variability across the batch where some product in vials will dry faster than others. It is difficult to eliminate the shelf temperature non-uniformity. However, measures can be taken to study the individual effects and minimize the degree to which they affect the shelf temperature uniformity.

The shelf uniformity is affected by several design factors such as fluid circulation path, number of the diversions along the flow path (due to its serpentine or spiral nature), dimension of the length of the shelf compared to its breadth, and type of heat transfer fluid. These factors lead to different flow characteristics (laminar flow along the length of the flow path and turbulent flow at the diversions) and the corresponding heat transfer characteristics (higher heat transfer coefficient at the diversions).

One of the factors that can help in achieving shelf temperature uniformity is the *edge* design. The edge design is not only important for the rigidity of shelves but it also has a significant impact on the rate of achieving homogeneous temperature over the shelf. One of the contributors to the paper has investigated (see Fig. 11, FEM simulation) the effect of the edge design and optimized the standard shelf with respect to its geometry and flow area without influencing the required shelf stability.

The simulation shows that the optimized edge design has significantly improved the temperature homogeneity within a specific timeframe.

It is to be noted that this freeze dryer manufacturer uses shelves with fixed Support Rails for holding the vials in the desired position. As per other freeze dryer manufacturer analyses, the avg. ΔT_{pdt} difference recorded for the side

Transient Thermal

Temperature 79

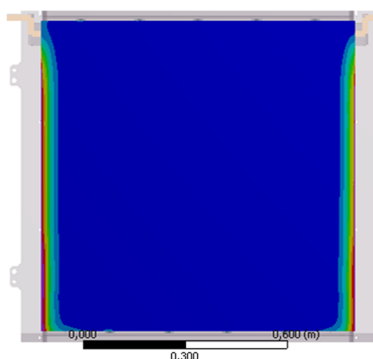
Type: Temperature

Unit: °C

Time: 5400

Max: -35.552

Min: -42.891



Shelf temperature distribution at vial area after 90 min.

Transient Thermal

Temperature 79

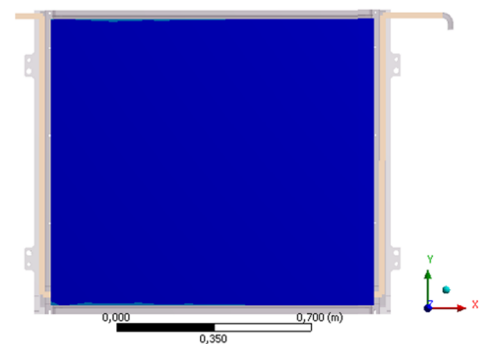
Type: Temperature

Unit: °C

Time: 5400

Max: -42.27

Min: -42.985



Shelf temperature distribution at vial area after 90 min with optimized edge design.

Fig. 11 Standard shelf design (left panel), optimized shelf design w.r.t. the edge material (right panel)

edge vials in contact with rails vs. no rails was 0.5°C. Thus, employing support rails might help minimizing the radiation effect from the sidewalls of the chamber.

According to one of the freeze dryer manufacturers, the chamber doors may have greater thickness compared to the walls to provide them with the required strength. The parts of the door region, in this case, are not insulated, whereas the sidewalls are thoroughly insulated. Therefore, it can be deduced that the temperatures in the chamber sides and door are likely to be non-uniform. As per this manufacturer’s analysis, the avg. ΔT_{pdt} between the edge vials exposed to the door vs. backside (exposed to duct) was 1.5°C.

Another manufacturer uses same wall thickness and reinforcement for the door and chamber wall with appropriate insulation. However, according to this manufacturer, there may be a discontinuity in radiated energy at the height of the nozzle for the slot door.

In any cases, the temperature sensors can be attached to the door/wall that serves to measure the warmest temperature in the chamber. This will help in identifying the largest edge effect during entire freeze drying process.

Accounting for Differences in Refrigeration Capacity and Dryer’s Performance

The refrigeration system is one of the most essential elements of a lyophilizer. It is crucial in maintaining chamber pressure and shelf temperature control during the process and is responsible in maintaining low temperatures on the condenser coils to drive water vapor from the chamber to the condenser [47]. Refrigeration systems on a freeze dryer may have different components (e.g., compressors vs. liquid

nitrogen heat exchangers), refrigerants (e.g., R-410A, R-507, Liquid Nitrogen), or condenser cooling mechanisms (e.g., direct expansion vs. heat transfer liquid cooled). Various factors determine what type of a refrigeration system is used on a freeze dryer. These include facility constraints such as the usable site space or the availability of utilities such as liquid nitrogen (LN2). During scale-up or tech transfer of a freeze drying process, it is highly recommended to understand what type of refrigeration system is present on each lyophilizer.

While compressor-based refrigeration systems have been used in freeze dryers for a long time, LN2-based systems have advanced in the last two decades. In addition, the presence of lesser moving parts in an LN2-based refrigeration system as opposed to a compressor-based refrigeration system will result in less downtime due to maintenance and repairs which may be a critical factor to consider during scale-up. Hence, given the choice between two lyophilizers, it may be preferable to transfer a lyophilization process to an LN2-based refrigeration system lyophilizer when all other factors are the same.

The condenser coil temperatures during the freeze drying process will influence process control. It is recommended to have predetermined condenser temperature set points during the process. In addition, having moderately higher condenser temperature set points such as -60°C will be advantageous for ice distribution on condenser coils as compared to extremely low set points such as -110°C [8, 47]. The type of refrigeration system will also influence the freezing process and affect the cooling rate of the shelves. It can be seen from Fig. 12 (left panel), for experiments performed on empty lyophilizer shelves that a liquid nitrogen-based refrigeration system will have a linear shelf cooling rate and will achieve typical ultimate freezing set points of -40°C or lower faster than a compressor-based system.

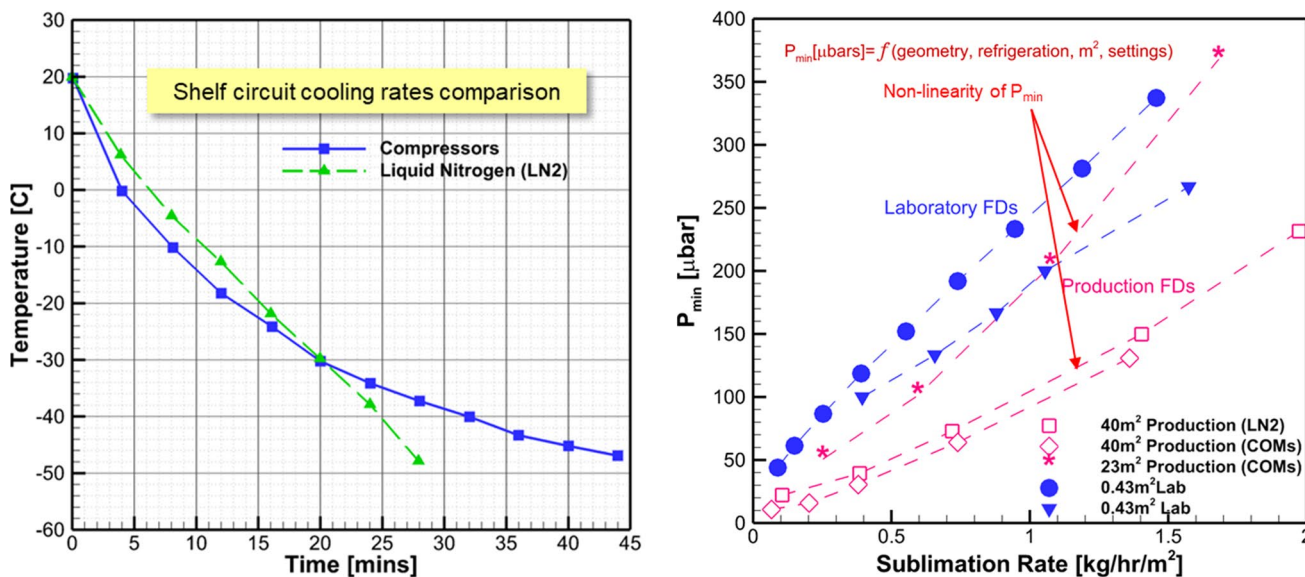


Fig. 12 Comparison of cooling rates between refrigeration and liquid nitrogen systems for the empty dryer (left panel). Comparison between equipment capability limits obtained for freeze dryers with different refrigeration systems (right panel)

The refrigeration system also impacts the freeze dryer equipment capability limit, a crucial component required for constructing design spaces. A comparison between equipment capability limits obtained for freeze dryers with different refrigeration systems is shown in Fig. 12 (*right panel*). The equipment capability limit for laboratory freeze dryers has a linear correlation between the sublimation rate and the minimum controllable chamber pressure. This implies that the equipment capability curve is solely the result of choked flow, and the refrigeration capacity poses no limitations. There is a non-linearity in the equipment capability curves of production freeze dryers due to limitations in their refrigeration capacity. Differences can also be seen in 40-m² production freeze dryers with liquid nitrogen and compressor-based refrigeration systems. Hence, it is recommended to not only characterize each freeze dryer with an equipment capability limit test, but also obtain the refrigeration capacity in terms of the total ice capacity of the condenser at operating set point temperatures.

There have been significant changes to the types of refrigerants being used in freeze dryers over the past several years. This is due to enhanced concerns regarding the environmental impact of refrigerants with respect to their global warming potentials (GWP) and ozone depletion potentials (ODP). For example, refrigerants R-410A, R-507, and LN2 have an ODP of 0, while a GWP of 2088, 3985, and 0 respectively. Alternate refrigerants such as air, carbon dioxide, or other hydrocarbon mixtures are being explored for use in freeze drying refrigeration systems. Only time will show whether any future developments can satisfy most user requirements. It is recommended that freeze drying experts and users be aware of the regulatory guidance, restrictions, and incentives in their respective regions while deciding on a particular refrigeration system for their lyophilizers.

The refrigeration system is one of the most crucial subsystems in a lyophilizer. The authors recommend that special care be given to understanding its type, capacity, and performance limits during the scale-up and tech transfer for any given process.

Accounting for Differences in Process Control

Chamber pressure is a critical parameter during primary drying. Pressure is measured and controlled by a variety of capacitance manometers, which measure pressures independent of the gas composition in the chamber. However, some older freeze dryers still use only a Pirani or similar pressure sensors reading of which depends on gas composition. In this case, adjustment of pressure set points [48] is needed to ensure that product temperature at commercial scale is similar to that in laboratory and remains below the collapse temperature. It is important to note that pressure sensors (capacitance manometers), used in commercial manufacturing, are regularly exposed to steam. Therefore, the baseline reading could shift by a few millitorr after each treatment. This potential impact should be accounted

for during a robustness study, preferably performed at the wide pressure design space. Since accuracy of capacitance manometers is linked to the full-scale pressure range (between 0.12 and 0.25% of full scale, MKS selection guide), 1 Torr head (0–1 Torr pressure range) is recommended for accurate pressure control, especially for low pressure (30–50 mT) cycles.

Pressure conversion during technical transfer is a common error (based on the primary S.I. units for the lyophilizer). It is important to review and check the pressure units while setting up the cycle and transferring it to the commercial-scale units. Basic pressure units are as follows: 1 atm = 760 Torr = 14.7 psia; 1 mTorr = 1.33 μ bar = 0.001 mbar.

Another factor is a proper proportional-integral-derivative (PID) control of shelf temperature, pressure, and condenser temperature. In laboratory dryers, pressure fluctuation is typically within ± 5 mTorr pressure range with the shelf temperature fluctuations within 2°C. The condenser temperature is typically very low (below –70°C) and has little effect on the process. In commercial dryers, pressure fluctuations could reach 30 mTorr which could force the evaluation of an extremely large pressure range design space during the robustness study. While shelf temperature fluctuations in commercial dryers do not typically exceed 5°C, condenser temperature (especially if condenser is cooled by direct expansion) could significantly vary causing fluctuations of condenser pressure and, in turn, chamber pressure. Proper PID control, adjusted for a load, could reduce impact of parameter variability on the product (refer to the section below). The PID controller parameters are typically set by the manufacturer prior to the factory acceptance test (FAT) for a clean, empty, and dry chamber. It is recommended that the user requirement specification (URS) should always include a sublimation test during the FAT so that PID refinement under a full load of water is executed by the equipment manufacturer. However, it is important to note that the PID settings tuned under a full-water load may need further refinement when formulations with markedly different sublimation rates are lyophilized in the same lyophilizer. In commercial operations, there is a significant difference in the gas composition due to which the constant values of PID may not be suitable for all stages of the drying process resulting in pressure and temperature fluctuations. One should note that pressure and shelf temperature fluctuations may change during the cycle (higher during the initial sublimation stage and lower toward the end of primary drying or vice versa). The authors suggest thorough testing should be performed during the site acceptance test (SAT) to ensure the dryer can handle a load without any fluctuations of temperature and pressure at the range and load (min and max sublimation rate) and the dryer is expected to operate. As a good practice, lyophilizers that routinely lyophilize formulations at extreme ends of the operational pressure range of a N₂-flow valve (e.g., MKS Type-248) will experience better pressure control during sublimation after the installation of separate N₂-flow valves in

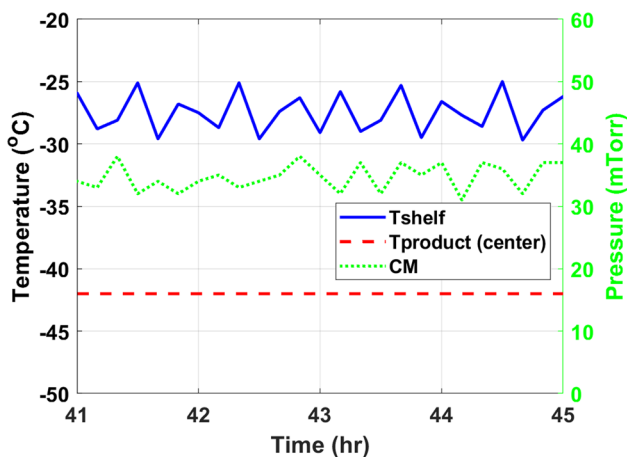


Fig. 13 The impact of short shelf temperature and pressure fluctuations on product temperature (note that values of shelf temperature and pressure were reported every 10 min, and actual fluctuation rates could be higher)

divergent loops, with each having its dedicated PID tuning. This recommendation also eliminates the need to frequently change the PID settings in such lyophilizers that are commonly employed by external contract manufacturing organizations (CMO). Ideally, one should have the ability to automatically adjust PID control inputs to maintain parameter fluctuations, such as pressure and shelf temperature, within a narrow range (± 5 mTorr and $\pm 3^\circ\text{C}$) during the entire lyophilization process. With advances in computational modeling and PAT technology, these designs are technically possible.

One of the big differences in process control between laboratory and commercial drying conditions is the ability to measure representative product temperature which is directly linked to the product quality. Due to introduction of automated loading systems, use of thermocouples or resistance temperature devices (RTD) is limited at commercial scale. Introduction of wireless sensors [48] combined with robotic sensor placement systems can allow product temperature measurements at defined locations.

Effect of Natural Process Parameter Fluctuations on Product Temperature

Fluctuations in pressure are expected during a freeze drying cycle. The challenge is determining the effect of the degree and duration of the fluctuation on product

temperature. In one example in a clinical size freeze dryer, fast fluctuations of pressure and shelf temperature did not significantly impact product temperature due to likely shelf-vial-product thermal inertia (Fig. 13).

The effect on the product temperature can be different for different products depending on their sensitivity to temperature changes and depending on the total solid content. Increasing the solid content increases the resistance to the mass flow of water vapor through the drying solid. Some companies may examine the effect of the degree and duration of a pressure fluctuation on product temperature using laboratory-scale studies. The studies may be useful if the design of the laboratory-scale freeze dryer is similar to the design of the full-scale freeze dryers. Laboratory-scale studies may also include examining the effect of the degree and duration of excursions in shelf temperature on the temperature of the product. An example of data collected from laboratory-scale studies is presented in Table III. The data in the table can be used to designate when conditions are outside of acceptable limits during full-scale manufacturing and determine when an investigation is needed. The time period for establishing when the process is outside of acceptable limits is determined based on the degree of the increase of product temperature when the excursion occurred for longer than 2 min.

Addressing the Impact of Container Closure Treatment

Commercially available containers for lyophilized products can vary in glass properties based on how they are manufactured as discussed above. Tubing glass is generally preferred for smaller volumes and invariably larger vials (50 or 100 mL) are made of molded glass. Compared to tubing glass, molded glass is more durable and heavier and, therefore, less susceptible to vials tipping over or breaking during manufacturing. But they also exhibit uneven thickness along the length of the vial and lower heat transfer capability. Tubing vials have greater heat transfer by contact conductivity through the areas that are in contact with the shelf, and greater gas conduction through the space between the shelf and the bottom of the vial than molded vials [49, 50]. The stippled bottom of the molded vials (Fig. 14) [51] in combination with the curvature at the bottom limits direct contact between the shelf surface area and vial bottom for heat transfer.

Table III Example of Data Collected at Laboratory Scale to Designate Acceptable Excursions in Shelf Temperature and Chamber Pressure During Full-Scale Freeze Drying

Process parameter	Limit (+/- from set point)	Time period designating outside of limits
Shelf temperature	Ramp: 4°C Dwell: 2°C	Duration longer than 2 min
Condenser temperature	$> 10^\circ\text{C}$ for set point of -70°C $> 5^\circ\text{C}$ for set point of -60°C	
Chamber pressure	Ramp: NMT 25 mT Dwell: NMT 25 mT	

Studies were conducted to compare heat transfer coefficients (K_v) of 50-mL tubing and molded vials (Table IV). The results indicated a significant difference in K_v when cycles were conducted at 100 mTorr and -10°C (Table IV). This discrepancy in K_v needs to be considered during cycle development if a vial change is necessitated.

Other than the vial types, the treatment of the vial also can have an impact on the process or the lyophilization cycle. The authors [33] demonstrated for borosilicate vials that vial washing and depyrogenation generate pits in the glass that impacts the glass surface properties. That could possibly contribute to vial fogging, a phenomenon which is not very well understood at commercial manufacturing based on the interaction of the formulation composition and the glass surface. Another treatment of the vial is related to the coating or composition of the vial itself. Exterior vial coating with polymers can help protect vials from damage due to shipping, surface pitting, and delamination caused by washing and depyrogenation and commercial-scale filling operations without significantly impacting heat transfer coefficients (unpublished data). The interior coating of the vial or the siliconization treatment can drastically reduce vial fogging. However, it is not preferred to proceed with siliconization due to potential challenges it can impose impacting the clarity of the solution for which siliconization assessment should be performed.

It is recommended to.

- Use tubing vials for uniform shelf contact and less variability in heat transfer.
- To measure the heat transfer coefficient when scaling up or using an alternate vial as there might be adjustments needed to the lyophilization cycle.

Example: A difference in K_v for the vial has the potential to impact the appearance of the dried product.

- Use processed glass to perform lab-scale experiments to match commercial treatment of the vials as closely as

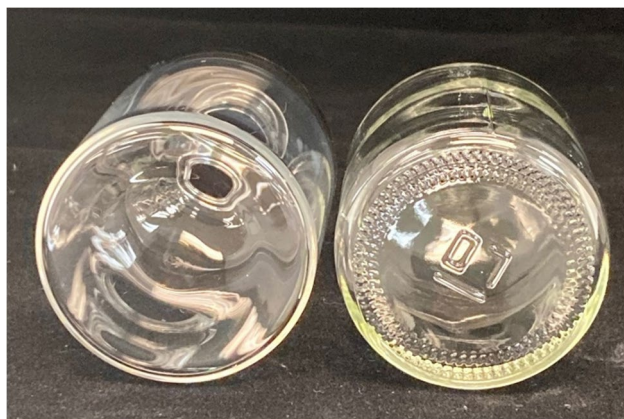


Fig. 14 Smooth bottom of tubing vial (left) vs. stippled bottom of molded vial (right)

Table IV Comparison of K_v for Tubing and Molded Vials

Glass	Batch average K_v ($\text{J/h}\cdot\text{cm}^2\cdot^\circ\text{C}$)
Molded	4.20
Tubing	5.99

possible. Glass can weaken after rinsing with water for injection and depyrogenation. This can lead to breakage of glass during the process.

- Treat stoppers in laboratory development process similarly to that in commercial manufacturing (targeting the same residual moisture).

Regulatory Aspects of Lyophilization Process Scale-up and Transfer

Knowledge and understanding of the regulatory expectations during the development, scale-up/validation, and manufacturing are critical to successful approval of NDA/BLA or MAA without receiving plethora of Information Requested (IR) and questions on deficiencies. An attempt is made here to bring to the attention and inform the process engineers involved in the design and development of the lyophilization process, the regulatory expectations, and the most common deficiencies observed in the regulatory submissions for lyophilized injectable products. The following information is extracted from publications and presentations made by the FDA staff.

It is expected that the applicant should clearly provide details on how the product has been developed and how development data and risk assessment were utilized to mitigate any risk associated with the scale-up and commercial manufacturing process. It is highly expected that a statistically sound sampling plan be designed and used during process validation to demonstrate drying uniformity across the self and between the shelves. The sampling plan should include both edge and center vials and testing should include physical quality (appearance, moisture content, and reconstitution time) and chemical quality attributes (stability indicating). If no scale-up is planned for commercial manufacturing, results of batch uniformity studies should be provided from the primary stability batches.

When determining the batch size of a drug product for primary stability batches (registration batches), the stability batch should be sufficient to allow for process capability to be established.

The minimum stability batch size required is discussed in the question and answer document pertaining to the 2014 FDA guidance for industry [52]. The inability to meet minimum batch size expectations during execution of the stability batches is one of the common deficiencies observed in

regulatory submissions. The 2014 FDA guidance for industry also provides specific batch size recommendations that are linked to target vial fill volumes, and any deviations from this guidance will call for adequate justification.

Since several PAT and process monitoring instrumentation are available now to monitor and control critical process parameters during the lyophilization [48, 53], the health authorities strongly expect development and implementation of process control strategy.

Some examples of common deficiencies related to lyophilization observed in the regulatory submissions listed below are extracted from reference [54].

In one case, the applicant intended to use the lyophilization cycle recipe used to manufacture of the exhibit batches for commercial manufacturing regardless of batch size. Therefore, the regulators asked the applicant to address the following points pertaining to scale-up plan for lyophilization.

- The critical process parameters of lyophilization (e.g., shelf temperature, ramp rate, and chamber pressure) should be adjusted to ensure the product has the same thermal history and quality attributes as the batch size increases for commercial production. If changes are not proposed, provide a scientific rationale for the adequacy of the established lyophilization cycle suitable for commercial production.
- For the proposed lyophilizers, provide the detailed information regarding the number of vials per tray, the number of trays per shelf, and total shelves per chamber to be utilized during scale-up for both exhibit and commercial batches.
- Demonstrate whether adequate control is put in place to determine the endpoint of primary drying during scale-up.
- Given that the dryer load condition is an important process variable during lyophilization process development and scale-up, explain how the risk associated with potential dryer overload (i.e., choked flow, condenser overload) is mitigated in commercial production based on proposed dryer load conditions.
- Provide a detailed sampling plan for the intended commercial batches to adequately capture the variation or uniformity of vials in different locations on each shelf with respect to the drug product critical quality attributes. In addition, revise the master batch record by including the proposed sampling plan accordingly.

In another case, the regulators asked the applicant to provide additional information on the lyophilization process development designed for the proposed drug product. It is desirable to understand how critical process parameters, such as shelf temperature and chamber pressure, were established. The regulators specifically asked the applicant to address the following points related to the characterization of the proposed lyophilization process:

- Explain how the shelf temperature used during primary drying was determined with respect to the glass transition temperature (T_g') and the collapse temperature (T_c). Include any thermal analysis data to support the rationale.
- Provide the drying charts exhibiting shelf temperature and chamber pressure continually recorded during the proposed lyophilization process for each manufactured exhibit batch. In addition, provide pressure rise data to justify the rationale for determining the end point of both primary and secondary drying. If primary drying endpoint is based on time, justify the duration.
- Provide detailed information regarding the number of vials per tray, the number of trays per shelf, and total shelves per chamber utilized during development studies of the exhibit batches. Provide the same information for the proposed commercial batches.
- Provide a detailed sampling plan for the commercial batches to adequately capture the variation or uniformity of vials in different locations on each shelf with respect to the drug product critical quality attributes.
- It is noted that annealing step is not included in the proposed lyophilization cycle although crystallization of mannitol during freezing is critical to minimizing the risk of vial breakage. Comment on what controls, if any, have been put into place to mitigate the risk associated with vial breakage during lyophilization and storage.

Demonstration of Functional Equivalencies Between Two Lyophilizers Made by Two Different Vendors

If you intend to leverage information and data generated to support the currently approved lyophilization cycle for the control of the new lyophilizers, provide information and data to demonstrate that these lyophilizers are “functionally equivalent”. This should include a comparison of the equipment and operational performance (e.g., from equipment qualification) of the currently approved and the new lyophilizers.

Equipment details provided should include, but not be limited to, the chamber dimensions and configurations as well as the condenser configuration and capacity. Operational performance comparisons provided should include, but not be limited to, the shelf area, shelf usage, maximum condenser load, empty shelf temperature uniformity, vacuum control, heating/cooling ramp rates, lowest condenser temperature, and minimum shelf temperature.

Demonstration of Performance Equivalence Between Two Same Type Lyophilizers Made by Same Vendor

Include information and data to support an evaluation of lyophilization process consistency for each new freeze dryer. Details should include, but not be limited to, the evaluation

of results from the lyophilization cycle operational parameters for each process validation lot as well as the evaluation of all relevant product quality attributes taken from select sample vials distributed across each shelf (i.e., center and edge positions) and throughout the freeze dryer (i.e., top, middle, and bottom shelves).

Recent Regulatory Guidance on Comparability of Freeze Dryers

The new version of the EU GMP Annex 1 from 2022 (EudraLex Volume 4) provides some more requirements on comparability of freeze dryers, including requesting to specify and document the loading pattern within the lyophilizer. In addition, samples for sterility testing are to be taken from different lyophilization loads. In case the manufacturing process results in sub-batches (e.g., when using not fully functionally equivalent freeze dryers), a sterility test needs to be performed separately for each sub-batch, and this approach may also be required for other finished product tests.

Other Considerations

Effect of Product Load on Pressure Control and Problem Mitigation

Full-scale production was planned for transfer to a third-party manufacturer with lyophilizers of similar design to the laboratory-scale lyophilizers at one large pharmaceutical company. A conservative approach was chosen for transfer of the lyophilization cycle with primary drying conditions conducted using a shelf temperature of +10°C with chamber pressure of 100 mTorr.

The in-process data for the initial cycles appeared with high variable pressure data for a 9-L batch and for a 35-L placebo batch (Fig. 15, top left and right panels, respectively).

The appearance of the pressure data from the capacitance manometer, Pirani gauge, and condenser pressure initially raised concerns of choked flow that dissipated later during primary drying. A 30-L batch was later prepared after examination and tuning of controls, valves, and sensors (Fig. 15, bottom left panel). A sawtooth pattern was still observed in the capacitance manometer and Pirani gauge pressure data.

Discussions were conducted with the manufacturer of the lyophilizer and an additional investigation was conducted to examine the control systems. The investigation found that the nitrogen control valve required tuning to balance the flow of nitrogen into the chamber as water vapor was removed and transferred to the condenser. The control of pressure within the lyophilizer was improved after tuning the flow of nitrogen (Fig. 8, bottom right panel).

In another example, it was determined that the default tuning parameters of this nitrogen bleed in valve were appropriate for a crystalline drug product cycle run. However, upon loading a different amorphous formulation with a different excipient percentage, the sublimation load was insufficient and caused larger pressure fluctuations. After eliminating the possibility of physical damage of the N2 flow valve through visual inspection, the valve PID tuning parameters were fine tuned during an engineering run resulting in an acceptable pressure profile.

Choice of Vial Internal Pressure After Lyophilization

The choice of head space pressure after lyophilization is complete has not been widely discussed in the literature. In our internal review, authors reported ranges between 0.3 and 0.83 Bar, 100 μBar and 0.8 Bar, or 0.8 and 0.95 Bar or full vacuum. Most often, 0.8 Bar pressure is used during stoppering. Note that all these residual pressure values were based on validated processes accompanied with successful CCIT performed. In some cases, high vacuum in a vial after the lyophilization process is needed to assist with faster reconstitution. In some cases, stoppers stick to the surface of the shelf during stoppering resulting in potential separation of stopper from the vial under gravity (only weight of vial and product are accounted). Attempts were made to determine the vacuum needed to seal vials without stoppers sticking to the shelf above. Figure 16 shows the minimal pressure gradient needed to maintain integrity of container closure after stoppering was complete and the shelves were raised to initiate unloading. The pressure gradient between atmospheric pressure and the pressure inside vial was calculated using Eq. 1 for 2R, 6R, 10R, 20R, 50R, and 100R vials (made by Schott).

$$\Delta P = \frac{(m_{vial} + m_{dry_product})}{\frac{\pi}{4}(D_{out}^2 - D_{in}^2)} g \quad (4)$$

Nominal fill volume values (2 mL for 2R vials, etc.) were used for calculations. Data show that even for 100R vials, in the worst-case scenario (30% solids), commonly used pressure value of 0.8 Bar during stoppering should be sufficient to maintain integrity of container closure. However, these calculations were made based on the assumption that the vial and stopper are compatible and there is no pop-up force contributing toward separation of vial and stopper. The recommendation is to use values in Fig. 16 as a guidance and use CCIT to ensure the integrity of container closure during validation process. The barometric pressure during stoppering should also be taken in account.

Besides partial vacuum inside vial after stoppering, sticking of stoppers can be managed by stoppering pressure and

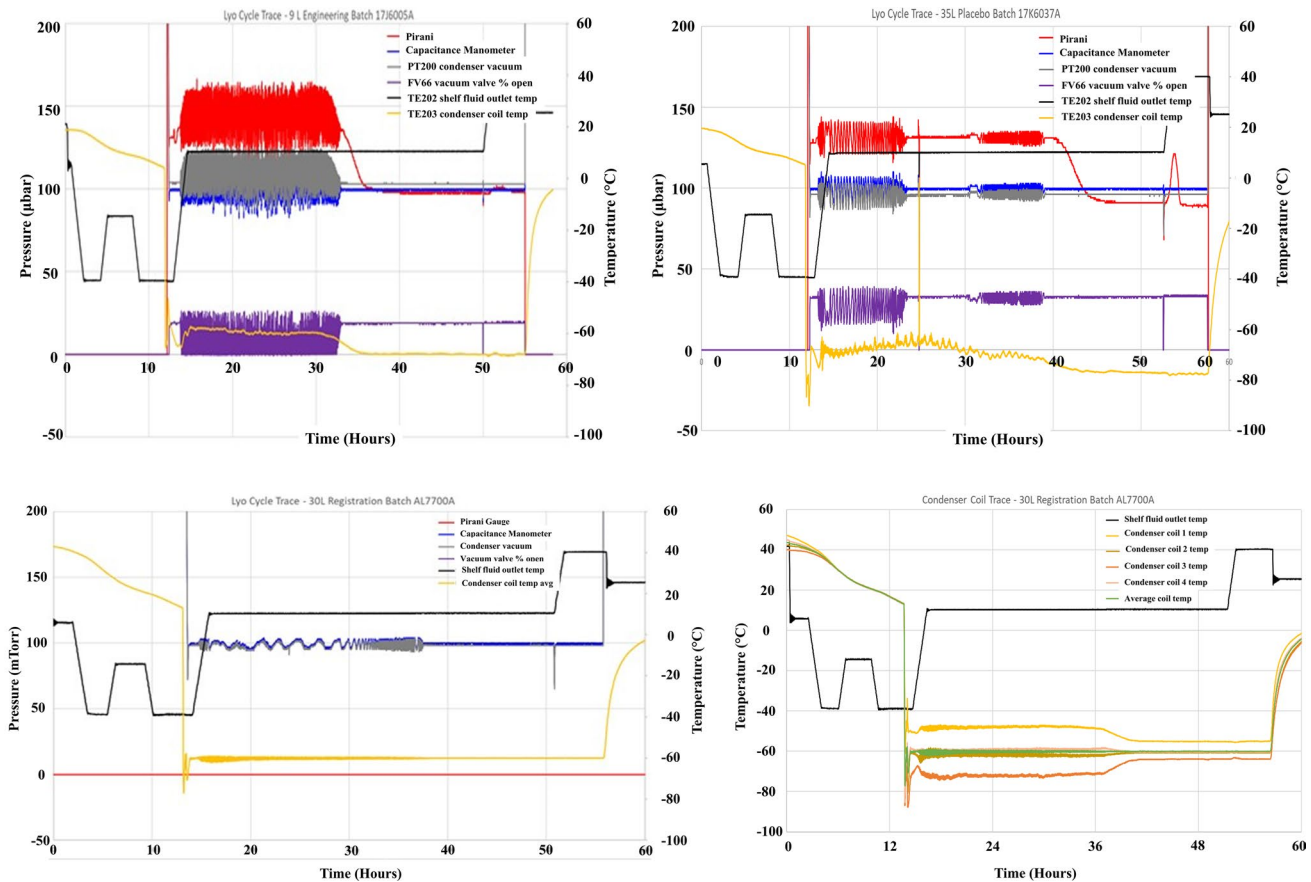


Fig. 15 Top left panel: in-process data for a 9-L engineering batch showing high variability in pressure during primary drying. Top right panel: in-process data for a 35-L placebo batch showing high variability in pressure during primary drying. Bottom left panel: in-

process lyophilization cycle data for a 30-L batch showing atypical pattern for capacitance manometer and Pirani gauge data. Bottom right panel: in-process lyophilization cycle data for a 30-L registration batch after tuning of the nitrogen control valve

duration of stoppering (specific to stopper material, compatibility between vial and stopper, etc.).

Future Direction

Application of CFD modeling to lyophilizer characterization will help to identify the limitations of freeze dryers and will allow generation of input parameters into the primary drying model. Real-time CFD modeling should, in principle, enable closed loop control based on critical process parameters (by controlling product temperature) for real-time optimization as well as assist in design of dryers which can withstand very high (about 1.5 kg/sq.m/hr) sublimation rates. Additional efforts need to be made to update the primary/secondary drying and freezing models to include syringes and cartridges and to also include elements of non-steady-state behavior. Obtaining alignment between the industry, academia, and regulatory

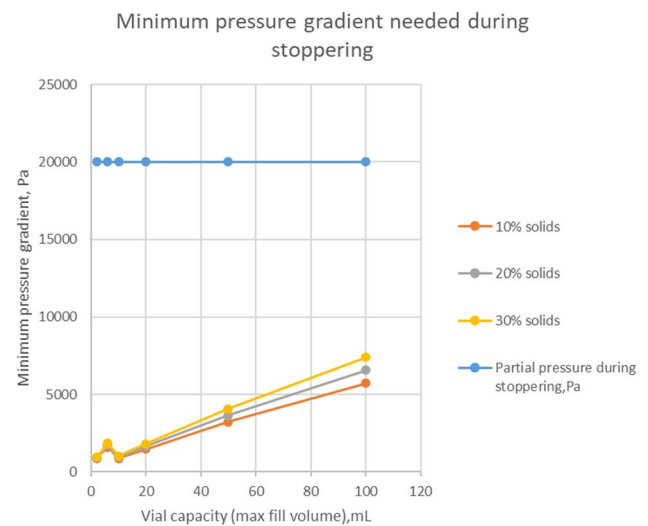


Fig. 16 A minimal pressure gradient across stopper needed to maintain integrity of container closure after stoppering

bodies should ensure approach to scale up and tech transfer of lyophilization process.

Recommended Best Practices for Scale-up and Technology Transfer in Freeze Drying

1. Follow the principle of rational process scale-up: Keep product below critical temperature while maintaining essentially the *same product temperature history* between lab-scale and commercial-scale dryers (within $\pm 1^\circ\text{C}$) which, in principle, should ensure identical product thermal history and product attributes, including stability.
2. Characterize the formulation and lyophilized products, and then demonstrate comparability of product quality attributes at both laboratory and production scale.
 - a. Successful scale-up requires a formulation that is characterized by thermal analysis and one that contains components amenable to freeze drying. A particular concern is formulations that have a very low maximum allowable product temperature during primary drying. Formulations with critical product temperatures of approximately -35°C or colder will be less successful during scale-up. It may be challenging to maintain low product temperatures for vials located on the edges of a shelf and avoid partial collapse.
 - b. Dried product from laboratory and commercial scale should be characterized by thermal analysis and compared. Amorphous solids should be examined using high temperature DSC to determine the Tg. It is generally useful to measure Tg as a function of residual moisture content in the region of the anticipated residual moisture specification during initial stage of development and storage. X-ray powder diffraction should be used to characterize the physical state of dried crystalline and semi-crystalline materials and to monitor them for changes over time.
3. Understand your equipment and process by key equipment parameter measurement and process analytical techniques (PAT) tools.
 - a. Any freeze dryer has a maximum sublimation rate that it will support. Minimum controllable pressure measurement is a relatively easy way to do this at any scale. Other aspects of equipment performance might be the lowest attainable shelf temperature and maximum shelf temperature ramp rate under load.
 - b. Use PAT tools (commonly accepted tools include comparative pressure measurement, tunable diode laser absorption spectroscopy (TDLAS), heat flux sensor, mass spec, nitrogen flow rate). Use appropriate process analytical techniques at both laboratory and production scale. Comparative pressure measurement is sensitive, robust, relatively inexpensive, and not dependent on monitoring of individual vials.
4. Match equipment between lab and production scale whenever possible. If possible, ensure that laboratory equipment is similar in design and construction to manufacturing scale equipment. For example, plexiglass doors are common on laboratory equipment, but manufacturing scale freeze dryers usually have stainless steel doors. Keep in mind that the higher thermal emissivity of plexiglass relative to stainless steel will accentuate the front edge effect in the laboratory-scale equipment.
5. Design and execute meaningful development and engineering runs.
 - a. Limited availability of API is a common problem in process development, and a development scientist may be tempted to use only a small number of vials. The problem is that the entire array of vials may be subject to the “edge effect,” resulting in misleading data on cycle time. Therefore, it is always a good practice to use a suitable (ideally matching thermal properties and similar cake resistance) placebo formulation with representative load making sure that the vials containing the API are well away (at least three vials) from the edge of the array.
 - b. Perform one or more engineering runs, perform moisture mapping study, and use NIR for 100% inspection. Conduct residual moisture mapping studies to establish acceptable secondary drying conditions.
6. Additional recommendations
 - a. Pay attention to details of stopper processing at both laboratory and production scale. The main concern is the potential for moisture migration from stopper to product during storage. Note that potential for moisture migration from stopper would be more critical for products with low solid content (typically for less than 30 mg cakes). Therefore, for product stability studies at the laboratory scale, it is a good practice to use stoppers that were processed targeting stopper residual moisture content similar to that in commercial samples. Also try to treat vials used in development studies similar to commercial vials (same depyrogenation cycle, etc.).
 - b. Utilize a backfill pressure which ensures a negative pressure in the vial head space.
 - c. Use of modeling is recommended accounting for differences in the lab and commercial-scale dryers (accounting for differences in ice nucleation temperature, cake resistance, and specific surface area).

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