AAPS PharmSciTech 2006; 7 (4) Article 93 (http://www.aapspharmscitech.org).

Evaluation of Manometric Temperature Measurement, a Process Analytical Technology Tool for Freeze-drying: Part II Measurement of Dry-layer Resistance

Submitted: December 22, 2005; Accepted: June 5, 2006; Published: December 1, 2006

Xiaolin Charlie Tang,[1,3] Steven L. Nail,[2,4] and Michael J. Pikal[1]

[1]School of Pharmacy, University of Connecticut, Storrs, CT
[2]Department of Industrial and Physical Pharmacy, Purdue University, West Lafayette, IN
[3]Current address: Formulation Development, Centocor Inc, Radnor, PA
[4]Current address: Lilly Research Laboratories, Eli Lilly & Co, Indianapolis, IN

ABSTRACT

The purpose of this work was to study the factors that may cause systematic errors in the manometric temperature measurement (MTM) procedure used to determine product dry-layer resistance to vapor flow. Product temperature and dry-layer resistance were obtained using MTM software installed on a laboratory freeze-dryer. The MTM resistance values were compared with the resistance values obtained using the "vial method." The product dry-layer resistances obtained by MTM, assuming fixed temperature difference (ΔT; 2°C), were lower than the actual values, especially when the product temperatures and sublimation rates were low, but with ΔT determined from the pressure rise data, more accurate results were obtained. MTM resistance values were generally lower than the values obtained with the vial method, particularly whenever freeze-drying was conducted under conditions that produced large variations in product temperature (ie, low shelf temperature, low chamber pressure, and without thermal shields). In an experiment designed to magnify temperature heterogeneity, MTM resistance values were much lower than the simple average of the product resistances. However, in experiments where product temperatures were homogenous, good agreement between MTM and "vial-method" resistances was obtained. The reason for the low MTM resistance problem is the fast vapor pressure rise from a few "warm" edge vials or vials with low resistance. With proper use of thermal shields, and the evaluation of ΔT from the data, MTM resistance data are accurate. Thus, the MTM method for determining dry-layer resistance is a useful tool for freeze-drying process analytical technology.

Corresponding Author: Michael J. Pikal, School of Pharmacy, U-2092, University of Connecticut, 372 Fairfield Road, Storrs, CT 06269-2092. Tel: (860) 486-3202; Fax: (860) 486-4998; E-mail: pikal@uconnvm.uconn.edu

KEYWORDS: Manometric temperature measurement, process analytical technology, freeze-drying, dry-layer resistance, product temperature, heterogeneity.

INTRODUCTION

Freeze-drying is widely used with pharmaceuticals to improve the long-term storage stability of labile drugs.[1,2] For given freeze-drying conditions (defined shelf temperature, chamber pressure and vials), the product temperature (T_p) in primary drying is determined by product dry-layer resistance ($\hat{R}_p$) with a high $\hat{R}_p$ yielding high product temperature and vice versa.[3] The T_p of a formulation with very low $\hat{R}_p$ (solute content less than 1%) is determined more by chamber pressure and less by shelf temperature. The T_p of a high product resistance formulation is more sensitive to shelf temperature change.[3] At a given product temperature, the ice sublimation rate is smaller at higher product dry-layer resistance. Typically, there is an increase of several degrees in T_p during primary drying, which is a result of the resistance increase with increasing dry layer thickness. Sometimes, the resistance change during primary drying reveals important information about the properties of the dried solid. For example, a dry-layer resistance that decreases or remains constant as dry layer thickness increases might indicate microcollapse during primary drying.[4,5] The product resistance also changes with freezing history. Usually, annealing the samples at a temperature higher than its glass transition temperature of a maximally freeze-concentrated solution (T_g'), decreases the product dry-layer resistance, thereby yielding shorter primary drying.[6,7] However, in one case, annealing during freezing increased the dry-layer resistance because of complications introduced by crystallization and, hence, increased the primary drying time.[8] Therefore, product dry-layer resistance can serve as a diagnostic tool and is a very important process analytical parameter for the freeze-drying process design and control.

Several methods have been reported for measuring product dry-layer resistance. A microbalance method, where freeze-drying is performed on samples in glass capillary tubes, needs specially designed devices and a well-calibrated temperature

sensor. The sublimation rate is directly calculated by mass loss as a function of time, and vapor pressure of ice at the sublimation interface is determined from the calibrated thermocouple temperature response.[6] However, since freezing behavior in the capillary tube is different from in a vial (more supercooling), microbalance resistance data are not a quantitative representation of freeze-drying in a vial. Alternately, resistance data may be measured during freeze-drying in modified vials. In this case, the vial is modified to facilitate pressure measurement inside the vial. The stopper resistance, which is treated as a constant under specified freeze-drying conditions, is determined from the relationship of the total ice sublimed and the pressures inside and outside of the vial after freeze-drying is completed. The ice sublimation rate is determined essentially by using the stopper as a flow meter.[9,10] The flow is assumed to be essentially viscous flow, and the stopper mass transfer coefficient is then determined by integration, over all of primary drying, of the difference between the square of the pressure inside the vial and the square of the pressure outside the vial. Then, with the mass transfer coefficient determined, the mass transfer rate from the measured pressures inside and outside the vial can be evaluated from any point in primary drying. The vapor pressure of ice is estimated from product temperature measured by thermocouple corrected for the temperature difference across the frozen layer.[6,10] This method is called the "vial method." Because resistance data generated in this fashion are sensitive to small measurement errors in both pressure and sample temperature, the data obtained are subject to both significant systematic and significant random errors.[9] Errors arising from pressure measurement may be eliminated by a modification of the vial method, where vials are extracted from the freeze-dryer using a sample thief and weighing.[5] The same sensitivity to temperature errors remains. Finally, the manometric temperature measurement (MTM) procedure may also be used to measure the product dry-layer resistance.[4,11]

MTM is a procedure to measure the product temperature at the sublimation interface during primary drying by quickly isolating the freeze-drying chamber from the condenser for a short time and by subsequent analysis of the pressure rise during this period.[4] The profile of the MTM data, which is a vapor pressure rise as a function of time, consists of 2 phases (ie, a fast-rise phase, which is governed by dry-layer resistance, and a plateau phase, which is determined by T_p). Analysis of the pressure rise data yields both product temperature at the ice sublimation interface and product dry-layer resistance. Actually, MTM measures the sum of the stopper resistance and the resistance of the dry layer, but the stopper resistance is normally negligible. The MTM method has the advantage of requiring no human intervention to place temperature sensors in the vials, thus there is less opportunity for product contamination, which is a huge advantage in manufacturing. Further, the method is easy to use and yields product dry-layer resistance in real time. However, during early use of the technique, we found that, in many cases, the MTM dry-layer resistances are far lower than the actual values.

The purpose of this research is to determine the origin of the errors in dry-layer resistance determined by MTM method. The possible causes are analyzed, and MTM procedures are proposed that allow the problems to be circumvented.

Materials and Methods

Sucrose, glycine, and mannitol were purchased from Sigma (St Louis, MO) and used without further purification. All reagents were of analytical grade. All the vials used for freeze-drying were 5-mL serum tubing vials from Fisher Scientific (Hampton, NH) (with internal cross-section area of 2.91cm^2 and external cross-section area of 3.51cm^2) and the stoppers were 20-mm double vent from Sigma.

Freeze-drying

Freeze-drying was performed with an FTS Dura-Stop/Dura-Top freeze-drier (Kinetics, FTS Systems Inc, Stone Ridge, NY) with the manometric temperature measurement (MTM) software installed. The pressure gauge (MKS capacitance, MKS Instruments, Andover, MA) with a resolution of ±1 mTorr was calibrated against our "standard," an MKS Baratron type 690 high-accuracy absolute capacitance manometer from 0 to 2000 mTorr. All solutions were prepared by weight volume ratio (wt/vol). For all freeze-drying runs, 150 sample vials were loaded on the middle shelf of the freeze-dryer. Thermal shields or radiation shields were used for some experiments including empty (dummy) vials around sample vials to reduce heat transfer from the freeze-dryer chamber wall and the door, and aluminum foil was attached to the inside of the chamber door to reduce the radiation from the door and outside. In one experiment, a low thermal conductivity material (Kimwipe, by Kimtech, subsidiary of Kimberly-Clark, Roswell, GA) was placed under edge vials (4 layers of Kimwipe sheets for side vials and 6 layers of Kimwipe sheets for front vials) to reduce heat transfer from shelf to edge vials (front and side vials).

The freeze-drying cycles for 5% glycine and mannitol were as follows: (1) freezing: 1°C/min to 5°C hold for 30 minutes; 1°C/min to −25°C hold for 60 minutes; 1°C/min to −40°C hold for 60 minutes; and (2) primary drying: conditions were changed according to specific experimental design. Chamber pressure was from 60 to 120 mT as designed; shelf temperatures were ramped 1°C/min to assigned shelf temperature (from −30°C to 43°C) as required and held until primary drying was completed.

The freeze-drying cycles for 5% sucrose were as follows: (1) freezing: 1°C/min to 5°C hold for 30 minutes; 1°C/min to −40°C hold for 60 minutes; and (2) primary drying: chamber pressure 80 mT; shelf temperatures were ramped 1°C/min to the assigned shelf temperature and held until primary drying was completed.

Manometric Temperature Measurement

MTM measurements were made at one or one-half hour intervals during primary drying, and pressure data were collected at the rate of 4 points per second during the MTM measurement. Typically, the data were collected for 25 seconds. The MTM equation was fit to the data by nonlinear regression analysis by means of a software package (Microcal Origin, MicroCal LLC, Northampton, MA) using one of 2 variations. Either the temperature difference across the frozen layer (ΔT) was fixed at 2°C, as is conventional [3], or ΔT was determined by the pressure rise data as discussed in this section.

The MTM equation, which describes pressure rise in the freeze-drying chamber (P, Torr) as a function of valve closure time during MTM (t, seconds)[4] may be written as follows:

$$P(t) = \underbrace{Pice - (Pice - P0)\cdot \exp\left[-\left(\frac{3.461\cdot N\cdot Ap\cdot Ts}{V\cdot(\hat{R}_p+\hat{R}_s)}\right)\cdot t\right]}_{Term\ 1} + \underbrace{0.465\cdot Pice\cdot \Delta T\cdot\left[1-0.811\cdot\exp\left(\frac{0.114}{Lice}\cdot t\right)\right]}_{Term\ 2} + \underbrace{X\cdot t}_{Term\ 3} \quad (1)$$

where P_{ice} is the vapor pressure of ice at the sublimation interface (fit, Torr); P_0, chamber pressure (set, Torr); N is the total number of samples vials (known); A_p is the inner cross-section area of vials (known, cm^2); T_s is shelf temperature (set, °C); V is the freeze-drying chamber volume (known, liter); $\hat{R}_p + \hat{R}_s$ (or $\hat{R}_{ps}$) is the total area of normalized product and stopper resistance (fit); L_{ice} is the ice thickness (calculated, cm); ΔT is the temperature difference between ice sublimation interface and bottom of the vials (fixed value at 2°C or determined from the data); and X is a constant (fit, Torr/s). An expression for ΔT can also be calculated using steady-state heat and mass transfer equations.[9]

$$\Delta T = \frac{[24.7\cdot Lice\cdot(Pice-P0)/(\hat{R}_p+\hat{R}s)-0.0102\cdot Lice\cdot(Ts-T)]}{1-0.0102\cdot Lice} \quad (2)$$

where T is the product temperature at the ice sublimation interface (K), which is related to vapor pressure of ice at the sublimation interface by Equation 3.[10,12]

$$T = \frac{-6144.96}{\ln(Pice) - 24.01849} \quad (3)$$

MTM curve fits were performed by use of Equation 1, where ΔT is a fixed value (2°C), and by the combined use of Equations 1 and 2. The results from both methods are compared in this article.

The converged curve fit yields vapor pressure of ice (P_{ice}), from which product temperature (T) is calculated from Equation 3, and total resistance of stoppers and product dry layer ($\hat{R}_p + \hat{R}_s$) as well as the linear parameter X (Equation 1, term 3).[4,13] The application of the parameter X is beyond the scope of this article. The stopper resistance ($\hat{R}_s$) is a constant at constant pressure and is often negligibly low. Therefore, the total resistance ($\hat{R}_p + \hat{R}_s$) can normally be taken as the area normalized product dry-layer resistance ($\hat{R}_p$).[9] The reproducibility of the MTM method was evaluated by repeated freeze-drying of 5% glycine at a shelf temperature of –20ºC and a chamber pressure of 80 mT with thermal shields. The results from 4 replicate experiments indicated that the maximum variation in MTM dry-layer resistance values was within 5%.

Calculation of Dry-layer Thickness

The ice sublimation rate is calculated using Equation 4.[6,9]

$$\frac{dm}{dt} = Ap\cdot\frac{Pice-Pc}{\hat{R}_p} \quad (4)$$

where dm/dt is the ice sublimation rate (g/h per vial); A_p is the internal cross-section area of vials (cm^2); P_{ice} is the vapor pressure of ice at the temperature of sublimation surface, which is determined by MTM (Torr); P_c is the vapor pressure of freeze-drying chamber (Torr); and $\hat{R}_p$ is the dry-layer resistance (or sum of the dry-layer and stopper resistance when stopper resistance is significant) obtained by MTM (cm^{2}Torrhour/g). The mass of ice sublimation, $m(t)$, in grams is calculated by numerical integration of dm/dt over the primary drying time, t. The dry-layer thickness is then calculated by Equation 5.[6]

$$l(t) = \frac{m(t)}{\rho I Ap\varepsilon} \quad (5)$$

where, $l(t)$ is the dry-layer thickness (cm) at time t; $m(t)$ is the mass of ice sublimed at time t (g/vial); ρ_I is the density of ice (g/cm^3); and ε is the volume fraction of ice, which is ~0.97 for 5% glycine, sucrose, or mannitol.

Thermocouple Placement

The 28-gauge Copper-Constantan (T-type) thermocouple temperature gauges (Omega Engineering, Stamford, CT) with a resolution of ±1ºC were calibrated at 0ºC using ice water.

Table 1. Manometric Temperature Measurement Fit Results for 5% Glycine in the Middle of Primary Drying (no thermal shields)*

		Freeze-drying Conditions		MTM Fit Results		
System No.	ΔT Used	T_s(°C)	P_c	T_p(°C)	R	Calculated ΔT
1	Calculated	–30	60	–43.0	1.71	0.02
	2°C	–30	60	–43.5	1.13	
2	Calculated	–20	80	–37.0	1.26	0.34
	2°C	–20	80	–37.3	1.13	
3	Calculated	+43	120	–24.0	1.54	2.89
	2°C	+43	120	–23.7	1.61	

*ΔT indicates the temperature difference and was calculated by Equation 2; MTM, manometric temperature measurements; T_s, shelf temperature; P_c, chamber pressure; T_p, product temperature; and R, dry-layer resistance.

The thermocouple product temperatures were measured at different locations during freeze-drying, including edge vials (front and side vials), and internal vials and were placed in the middle of the vials touching the vial bottoms.

Results and Discussion

Effect of Evaluating Temperature Difference From the Data

The temperature difference between the ice sublimation interface and the bottom of the vials appears in the MTM equation (Equation 1), and this parameter can either be arbitrarily set to a constant value, such as 2°C,[4] or can be evaluated from the pressure-time curve as outlined in the section Manometric Temperature Measurement. The MTM equation (Equation 1) with ΔT = 2°C or the MTM equation with ΔT determined from the data (Equations 1 and 2) was fit to the MTM raw data collected under different freeze-drying conditions. The curve-fitting results from both methods are compared in Table 1. The values of ΔT obtained by Equation 2 are much smaller than 2°C (only 0.02°C and 0.34°C) at low shelf temperatures (−30°C and −20°C). At high shelf temperature (43°C) and chamber pressure (120 mTorr), the calculated ΔT value is greater than 2°C. For the −30°C shelf-temperature experiment, the MTM $\hat{R}_p$ obtained using Equation 2 to calculate ΔT was 1.7 cm^2Torrhour/g, while using a fixed value of 2°C for ΔT yielded a much smaller resistance value ($\hat{R}_p$ = 1.1 cm^2Torrhour/g). The $\hat{R}_p$ value of 1.7 cm^2Torrhour/g is more consistent with the $\hat{R}_p$ value from the vial method (Figure 1). Although the product temperatures obtained from the 2 methods are in satisfactory agreement (0.5°C difference), the difference in resistance values is significant. At higher shelf temperature (−20°C and +43°C) and higher chamber pressure (80 and 120 mTorr), agreement between MTM resistances is better. Thus, the errors introduced by assuming a constant ΔT are insignificant in temperature measurement but clearly may be large for resistance measurement particularly for slow freeze-drying at low temperature. Therefore, using Equation 2 for ΔT is clearly the best practice.

MTM Dry-layer Resistance ($\hat{R}_p$) Under Different Freeze-drying Conditions

The Effect of Shelf Temperature: No Thermal Shield

Two freeze-drying experiments of 5% glycine solution (150 vials, 2-mL fill in 5-mL tubing vials) were conducted under the same conditions except for different shelf temperatures (20°C and −20°C). No thermal shield was used for the MTM experiments. That is, neither empty vials around sample vials nor aluminum foil on the door were used. The dry-layer resistance values from MTM method were compared with those from vial method in Figure 1 (M. J. Pikal and S Shah, Eli Lilly and Co, unpublished observations 1995). The vial method is the standard method for measuring the dry-layer resistance and, with due consideration for the limited precision, such data are assumed accurate. Figure 1 shows that the dry-layer resistance values obtained by MTM are much smaller than those obtained by the vial method when the shelf temperature is low (−20°C). This low resistance problem was especially significant at the beginning of primary drying when the dry-layer resistance was low.

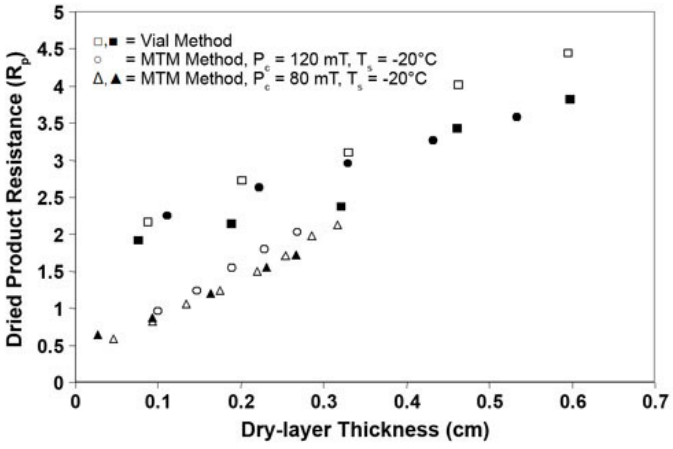

Figure 1. Product dry-layer resistance for 5% glycine: vial method (squares) compared with manometric temperature measurements (MTM) method at different shelf temperatures and pressures. R_p indicates product temperature; P_c, chamber pressure; and T_s, shelf temperature.

The MTM fitted results are consistent with vial method results when the same solution was freeze dried at high shelf temperature (20°C) (Figure 1). The product temperatures measured by MTM were consistent with thermocouple results for both shelf temperatures (data not shown).[14]

The Effect of Chamber Pressure: No Thermal Shields

Two different chamber pressures (80 mTorr and 120 mTorr) were used for 2 identical freeze-drying experiments under the same conditions (5% glycine, 150 vials, 2-mL fill in 5-mL tubing vials). The low shelf temperature of −20°C was used for both experiments. Figure 1 shows that MTM resistance values at both pressures were significantly lower than the resistance values obtained from the vial method, although the MTM results at higher chamber pressure (120 mTorr) appeared to be a little closer to the true resistance values.

The Effect of Thermal Shields

A row of empty vials (dummy vials) around sample vials and aluminum foil applied inside the freeze-drying chamber door were used as thermal shields. We have found that the use of thermal shields can reduce the product temperature heterogeneity between sample vials during primary drying.[15] Dry-layer resistance values obtained by the MTM method are were compared with corresponding values determined using the vial method in Figure 2 for glycine and mannitol, and in Figure 3 for sucrose. In Figure 2A, results for 5% glycine freeze-dried at a shelf temperature of −20°C, and a chamber pressure of 80 mTorr are compared. The MTM values obtained from freeze-drying without thermal shields were much lower than values obtained by the vial method. However, good agreement was found when thermal shields were used. Results were similar when 5% mannitol or 5% sucrose were freeze-dried (Figure 2B and Figure 3). Thus, we conclude that the MTM method for resistance measurement does yield accurate data, provided thermal shields are employed.

The Effect of Low Thermal Conductivity Material Under the Edge Vial

These experiments were designed to freeze-dry 5% glycine under the same conditions as mentioned before (150 vials, 2-mL fill in 5-mL tubing vials, shelf temperature of −20°C and chamber pressure of 80 mTorr and no thermal shields) except that low-thermal conductivity material (ie, Kimwipe tissue) was applied under edge vials (front and side vials) to reduce the heat transfer from the freeze-drying shelf to edge vials, so that the temperature difference between edge vials and internal vials was reduced. This procedure was at least partially successful. We note that the product temperature

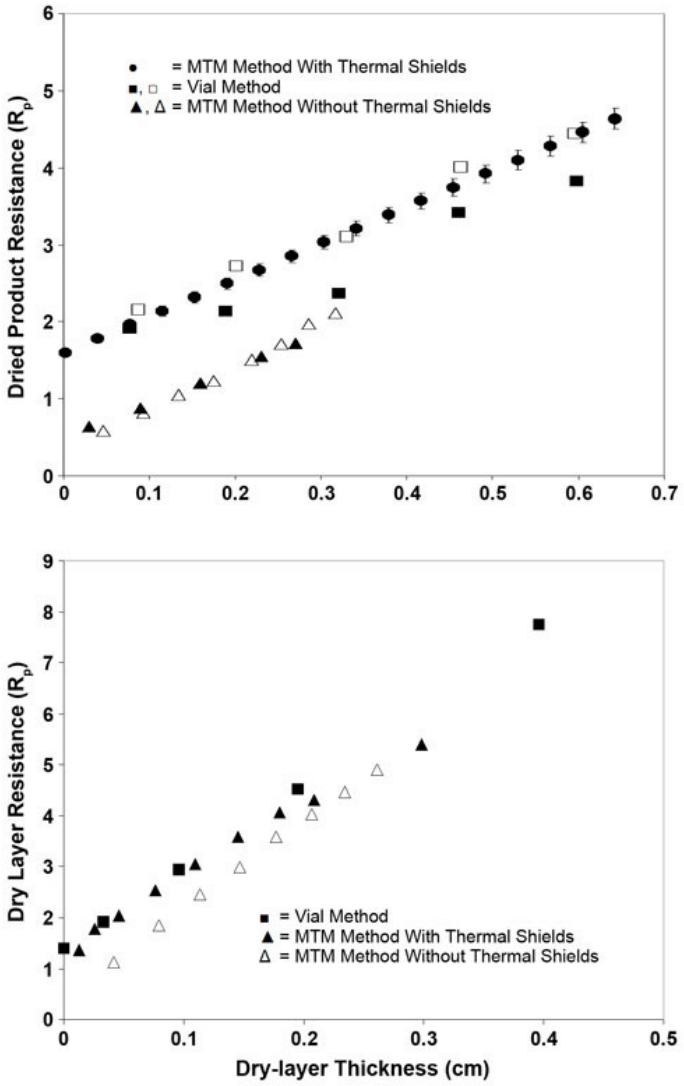

Figure 2. Product dry-layer resistance of 5% glycine (upper panel) and 5% mannitol (bottom panel) at shelf temperature of −20 ºC and chamber pressure of 80 mTorr: vial method (squares) compared with MTM method (ΔT evaluated from data). MTM indicates manometric temperature measurements (MTM); ΔT, temperature difference.

difference between vials was controlled within 2°C during primary drying, although the atypical radiation from chamber wall and door still exists (since no thermal shields are applied), compared with difference of 4°C between front vials and interior when all vials contact the shelf in the experiment (with no thermal shields). The MTM dry-layer resistances obtained in these series of experiments are compared with previous results in Figure 4. Contrary to results obtained with all vials in contact with the shelf, resistance results obtained in the "Kimwipe experiment" were *not* lower than vial method results. These results indicate that the low MTM resistance problem is not caused by the radiation heat transfer mechanism itself but rather arises from product temperature heterogeneity in the freeze-dryer. Of course, normally, the side radiation effect does cause the temperature heterogeneity.

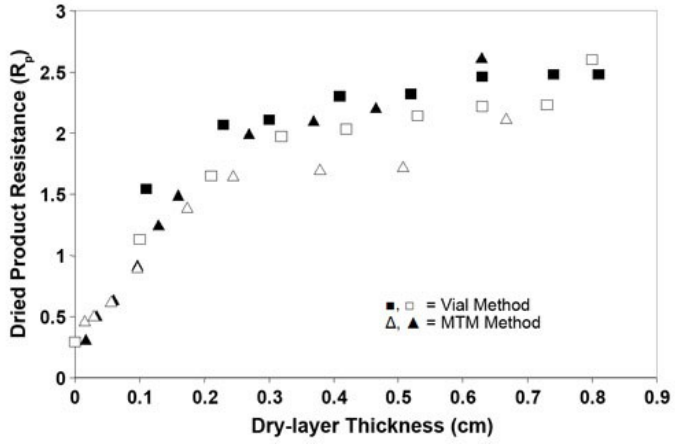

Figure 3. Products dry-layer resistance for 5% sucrose: vial method (squares) compared with MTM method (thermal shields applied and ΔT evaluated from data). MTM indicates manometric temperature measurements (MTM); ΔT, temperature difference.

The Effect of Mixed Samples

To provide a situation of extreme temperature heterogeneity, 138 vials of 5% glycine were freeze-dried with one row (12 vials) of pure water at the front. The freeze-drying was conducted at a shelf temperature of −20°C and chamber pressure of 80 mTorr with thermal shields applied. The MTM equation fit the pressure rise data well, but the MTM dry-layer resistance value obtained (0.79 Torr*h/g/cm^2) was less than one third of the actual resistance value measured by the vial method (2.61 Torr*h/g/cm^2) for 5% glycine.[9] The MTM product temperature (−38.6°C) was close to the pure water temperature (−39°C by thermocouple) and much lower than the product temperature of 5% glycine (−32°C by thermocouple) even though there were far more "warm" glycine vials than "cold" water vials (138 glycine vials compared with 12 water vials).[15] Thus, as noted elsewhere,[13]the MTM procedure measures an average temperature heavily weighted in favor of the coldest vials (ie, the pure water vials). The MTM dry layer resistance is not a simple average between different resistance values in a freeze-drying system. A few low resistance sample vials can dramatically change the MTM dry-layer resistance. Therefore freeze-drying a mixture of products with different dry-layer resistances is not recommended if measurement of the MTM resistance is intended.

Origin of MTM Dry-layer Resistance Variations With Freeze-drying Conditions

The tendency of the MTM procedure to yield low dry-layer resistance values is especially serious when freeze-drying is conducted under conditions of low shelf temperature, low chamber pressure, and without any thermal shields. The problem is minimized whenever the samples are freeze-dried at high shelf temperature (shelf temperature close to ambient), or at higher chamber pressure, or by use of thermal shields. All the aforesaid conditions for obtaining improved results suggest that radiation heat transfer to the vials at the edge of the vial array (denoted, "atypical radiation") is the reason for the low dry-layer resistance problem in MTM.[15] The heat transfer during primary drying is described by Equation 6.[9]

$$\frac{dQ}{dt} = Av \cdot Kv \cdot (Ts - Tb) \qquad (6)$$

where dQ/dt is heat transfer rate (cal/h per vial); A_v is vial cross-sectional area (cm^2); T_s is the shelf surface temperature (°C), T_b is the temperature of vial bottom (°C); and K_v is heat transfer coefficient of the vials (cal/s/K/cm^2), which consists of 3 components (Equation 7)[9]:

$$Kv = Kc + Kg + Kr \qquad (7)$$

where K_c is the contribution of heat transfer from direct contact between vials and shelf; K_g is the contribution of gas conductivity; and K_r is the contribution of radiation from shelf, and surroundings. Typically, K_c and K_r are constant, and K_g is a function of chamber pressure, increasing with chamber pressure.[16] Side radiation comes from the chamber wall or door directly onto edge vials, but not directly onto interior vials. The effect of side radiation is

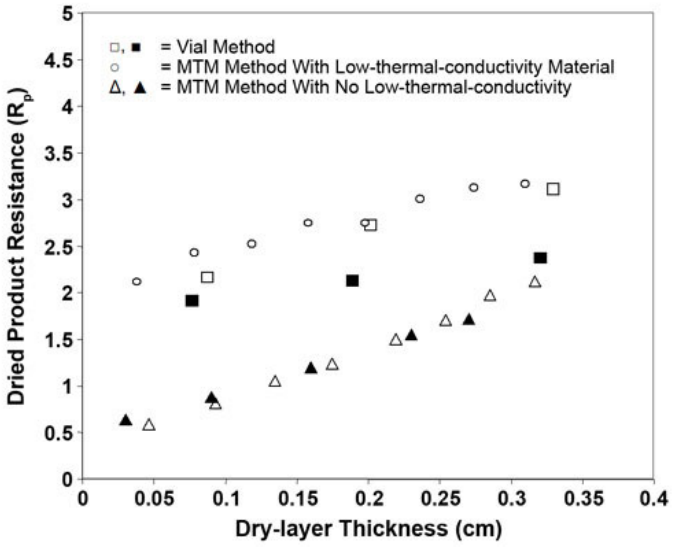

Figure 4. Product dry-layer resistance for 5% glycine: vial method (squares) compared with MTM method and the effect of low-thermal-conductivity material under the edge vials. MTM indicates manometric temperature measurements.

reduced if the shelf temperature is close to ambient temperature, because the fraction of total heat flow from the chamber wall and door is less when the total heat flow from the shelf is high, as with a shelf temperature near ambient. The fraction of total heat transfer arising from atypical radiation is decreased if a higher chamber pressure is used for freeze-drying, because the total heat transfer from the shelf by gas conductivity is increased, and the heat transfer by radiation does not change. The atypical radiation is directly reduced by use of thermal shields. Here, radiation heat transfer from the chamber wall is reduced by the surrounding empty vials, and heat transfer from the door is reduced by the attached low-emissivity aluminum foil. The fact that the accuracy of the MTM resistance data are improved whenever the fraction of heat transfer from atypical radiation decreases indicates that the atypical radiation heat transfer to the edge vials causes the MTM low resistance problem.

A Theoretical Analysis of the Effect of Temperature Heterogeneity

Typically, the product temperature of the front vials is highest, and that of the internal vials is lowest, and the temperature of the edge vials on the sides is in between but closer to the internal vials.[9,15] Here, we consider only the contribution of water vapor flow into the chamber from sublimation of ice at constant temperature; that is, we consider only "Term 1." We postulate a distribution of product temperatures that may be divided into 2 distinct classes (to acknowledge vial temperature heterogeneity in the simplest way): "hot" vials, labeled No. 1; and "cold vials," labeled No. 2. The "hot" vials would represent vials along the front edge, while the cold vials would represent all other vials. This representation is perhaps an oversimplification, but the qualitative results sought will not be adversely affected by this simplification. We consider, for the moment, that we may distinguish vapor originating from the different vial classes. Further, we assume that vapor from the "hot" vials will condense on the colder vials when the partial pressure of water coming from the "hot" vials approaches the dew point of the colder vials. The increase in pressure coming from the cold vials is written as usual in the form,

$$\frac{dP_2}{dt} = \left(\frac{dP_2}{dt}\right)_{\text{sublimation}} = N_2 \frac{RT}{MV} \cdot A_p \frac{(P20 - P)}{\hat{R}_{\text{ps}}} \qquad (8)$$

where $P20$ is the vapor pressure of ice at the sublimation interface of the cold vials (class 2) of number N_2, P_2 is the "partial" pressure of water from class 2 vials, and P is the chamber pressure. The other symbols have their usual meanings. The expression for increase in pressure coming from the "hot" vials is a bit more complex, containing a term similar to the right-hand side of Equation 8 describing sublimation but also containing a term describing condensation on the cold vials,

$$\begin{aligned}\frac{dP_1}{dt} &= \left(\frac{dP_1}{dt}\right)_{\text{sublimation}} + \left(\frac{dP_1}{dt}\right)_{\text{condensation}} \\ &= N_1 \frac{RT}{MV} \cdot A_{\text{p}} \frac{(P10 - P)}{\hat{R}_{\text{ps}}} - k \cdot P \end{aligned} \qquad (9)$$

where k is the "condensation constant," which may be evaluated by noting that, according to our assumptions, the value of dP_1/dt becomes zero at equilibrium when the chamber pressure is the vapor pressure of the cold vials; $P20$ is due to condensation. Thus, setting $dP_1/dt = 0$ when $P = P20$, we find the value of k is given by

$$k = N_1 \frac{RT}{MV} \cdot A_{\text{p}} \frac{(P10/P20 - 1)}{\hat{R}_{\text{ps}}} \qquad (10)$$

The total pressure rise is (assuming ideal gas behavior), $dP/dt = dP_1/dt + dP_2/dt$, which upon combination of Equations 8 through 10 may be written

$$\frac{dP}{dt} = N \cdot \frac{RT}{MV} \cdot \frac{A_{\text{p}}}{\hat{R}_{\text{ps}}} \cdot \left[(X_1 P10 + X_2 P20) - \left(X_1 \frac{P10}{P20} + X_2 \right) \cdot P \right] \qquad (11)$$

where $N = N_1 + N_2$, and X_1 is the fraction of vials of "type I." We now define P_{ice} as the number average vapor pressure of ice, $P_{ice} = X_1 P10 + X_2 P20$, and rearrange the differential equation, Equation 4, prior to integration to yield

$$\frac{dP}{dt} = N \cdot \frac{RT}{MV} \cdot \frac{A_{\text{p}}}{\hat{R}_{\text{ps}}} \cdot P_{ice}(1 - P/P20) \qquad (12)$$

Integration of Equation 12 subject to the initial condition, $P = P_{\text{c}}$ at $t = 0$, where P_{c} is the chamber pressure prior to the MTM test, then gives

$$\begin{aligned} P &= P20 - (P20 - P_c) \cdot \exp\left(-N \cdot \frac{RT}{MV} \cdot \frac{A_{\text{p}}}{\hat{R}_{\text{ps}}} \cdot \frac{P_{ice}}{P20} \cdot t \right) \\ &= P20 - (P20 - P_c) \cdot \exp(-B \cdot t) \end{aligned} \qquad (13)$$

where the symbol B has been used for the collection of terms that multiply time, t, inside the exponential.

Note that Equation 13 is exactly of the same form as term 1 of the original MTM equation. There are only 2 differences. First, consistent with our assumption of condensation of vapor from "hot" vials on the "cold" vials, the chamber pressure approaches the vapor pressure of ice in the cold vials at equilibrium, meaning that the MTM temperature obtained by a fit to

pressure rise data will give the temperature of the cold vials as the MTM temperature. Second, the term, B, now contains the ratio $P_{ice}/P20 = 1 + X_1 \cdot (P10/P20)$, which is greater than unity. Thus, resistance obtained by fitting the usual MTM equation to the data will yield a resistance that is too small by the factor $P_{ice}/P20$. This conclusion, based upon this theoretical model, is in agreement with our experimental observations, thus supporting our hypothesis that heterogeneous temperature is responsible for giving low resistance values.

The modified MTM equation (Equation 13) needs thermocouple temperature values to allow analysis of pressure rise data. The bottom line is that the modified MTM equations serve as a tool to explain the origin of low MTM resistance problem. However, the modified MTM equations are not well suited to the routine use of MTM simply because the whole point of MTM is to avoid use of thermocouples.

CONCLUSIONS

Accuracy in product dry-layer resistance measurement by MTM is improved if ΔT is evaluated from the pressure rise data rather than taken as a constant (2°C). The MTM dry-layer resistance values are usually lower than the actual values, especially when freeze-drying is performed at low shelf temperature and low chamber pressure. This problem is caused by product temperature heterogeneity in freeze-drying resulting from atypical radiation from the environment, but this problem can be solved or at least minimized whenever thermal shields are applied. Thus, with suitable precautions and modifications, the MTM method is a useful alternative tool for measuring dry-layer resistance.

ACKNOWLEDGMENTS

This project was sponsored by National Science Foundation, Center for Pharmaceutical Processing Research (NSF/CPPR).

REFERENCES

1. Pikal MJ. Lyophilization. In: Dekker M, ed. *Encyclopedia of Pharmaceutical Technology.* 2001:1299–1326.

2. Carpenter JF, Pikal MJ, Chang BS, Randolph TW. Rational design of stable lyophilized protein formulations: some practical advice. *Pharm Res.* 1997;14:969–975.

3. Tang X, Pikal MJ. Design of freeze-drying processes for pharmaceuticals: practical advice. *Pharm Res.* 2004;21:191–200.

4. Milton N, Pikal MJ, Roy ML, Nail SL. Evaluation of manometric temperature measurement as a method of monitoring product temperature during lyophilization. *PDA J Pharm Sci Technol.* 1997; 51:7–16.

5. Overcashier DE, Patapoff TW, Hsu CC. Lyophilization of protein formulations in vials: investigation of the relationship between resistance to vapor flow during primary drying and small-scale product collapse. *J Pharm Sci.* 1999;88:688–695.

6. Pikal MJ, Shah S, Senior D, Lang JE. Physical chemistry of freeze-drying: measurement of sublimation rates for frozen aqueous solutions by a microbalance technique. *J Pharm Sci.* 1983;72: 635–650.

7. Searles JA, Carpenter JF, Randolph TW. Annealing to optimize the primary drying rate, reduce freezing-induced drying rate heterogeneity, and determine Tg' in pharmaceutical lyophilization. *J Pharm Sci.* 2001;90:872–887.

8. Lu X, Pikal MJ. Freeze-drying of mannitol-trehalose-sodium chloride-based formulations: the impact of annealing on dry-layer resistance to mass transfer and cake structure. *Pharm Dev Technol.* 2004;9:85–95.

9. Pikal MJ, Roy ML, Shah S. Mass and heat transfer in vial freeze-drying of pharmaceuticals: role of the vial. *J Pharm Sci.* 1984;73:1224–1237.

10. Pikal MJ. Use of laboratory data in freeze drying process design: heat and mass transfer coefficients and the computer simulation of freeze drying. *J Parenter Sci Technol.* 1985;39:115–139.

11. Tang XC, Nail SL, Pikal MJ. Mass Transfer in Freeze Drying: Measurement of Dry-layer Resistance by a Non-Steady State Method (the MTM Procedure). Paper presented at: AAPS Annual Meeting; November 14-18; New Orleans, LA. 1999.

12. Jancso G, Pupezin J, Van Hook WA. The vapor pressure of ice between 0.01 and −100 C. *J Phys Chem.* 1970;74:2984–2989.

13. Chang L, Tang X, Pikal MJ, Milton N, Thomas L. *The Origin of Multiple Glass Transitions in Frozen Aqueous Solutions.* Proceeding NATAS Annual Conference Thermal Analysis and Applications. Bowling Green, NY, NATAS. 1999:624–628.

14. Shamblin SL, Tang X, Chang L, Hancock BC, Pikal MJ. Characterization of the time scales of molecular motion in pharmaceutically important glasses. *J Phys Chem B.* 1999;103: 4113–4121.

15. Tang X, Nail SL, Pikal MJ. Evaluation of manometric temperature measurement (MTM) in freeze drying. Part I: product temperature measurement [serial online]. *AAPS PharmSciTech.* 2006;7:E14.

16. Nail SL. The effect of chamber pressure on heat transfer in the freeze drying of parenteral solutions. *J Parenter Drug Assoc.* 1980;34:358–368.