DE MONTFORT UNIVERSITY LEICESTER

Freeze-drying process optimization using through vial impedance spectroscopy

EPSRC EHDA Network

Intl. Pharm. Tech. Conference, Leicester

Friday 4th November, 2016

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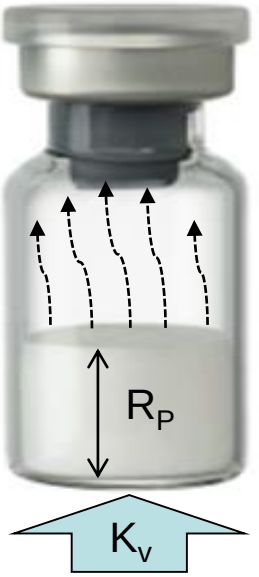
Leicester School of Pharmacy

BIOPHARMACEUTICAL QUALITY BY DESIGN

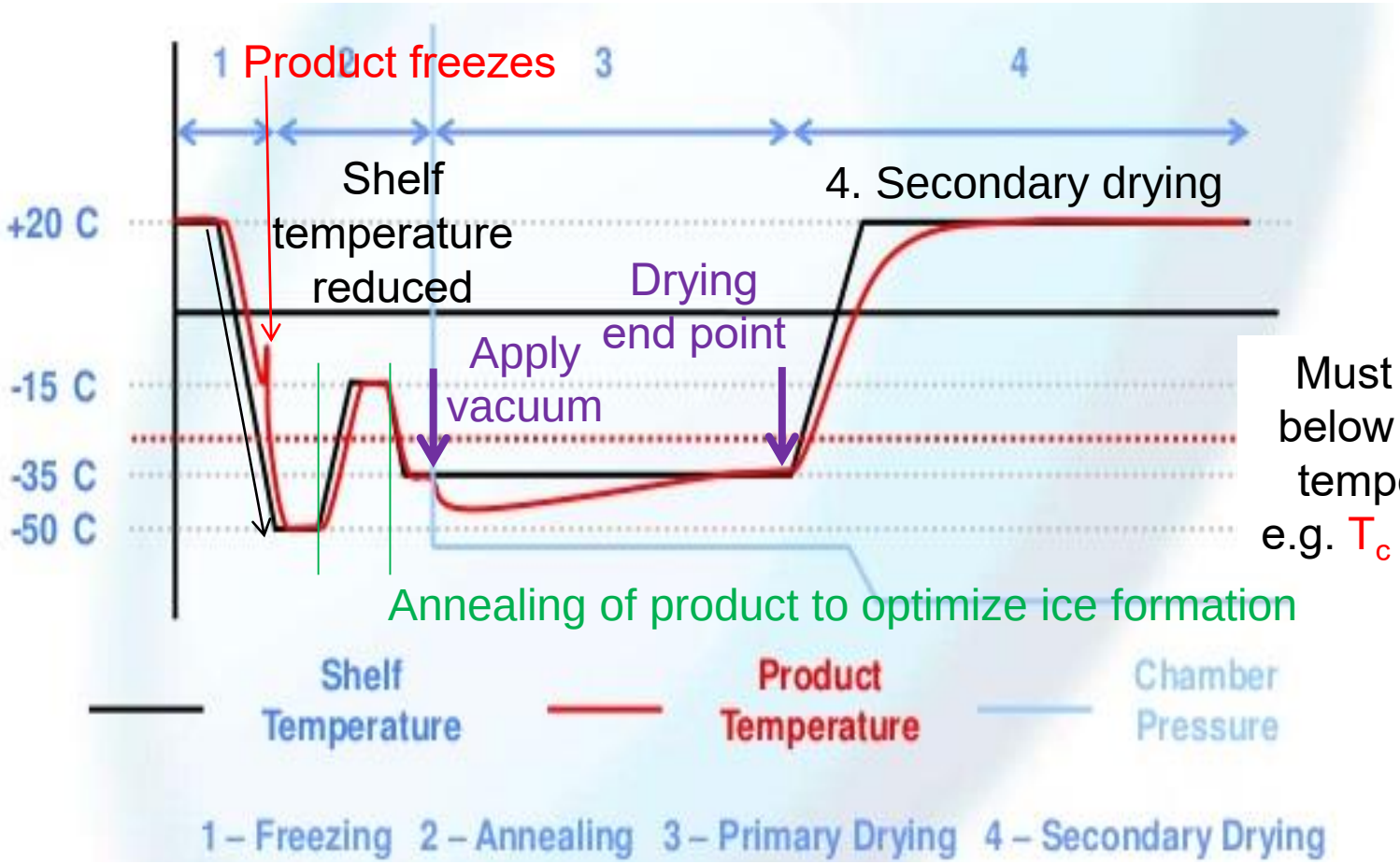
Outline

- Freeze-drying and the design space
- Introduction of Through Vial Impedance Spectroscopy (TVIS) Technology

- TVIS Applications for Determination of
  - Heat transfer coefficient through the vial base K_V
  - Dry layer resistance which impedes the of water vapour sublimation R_P



LYOPHILLIZATION STEPS



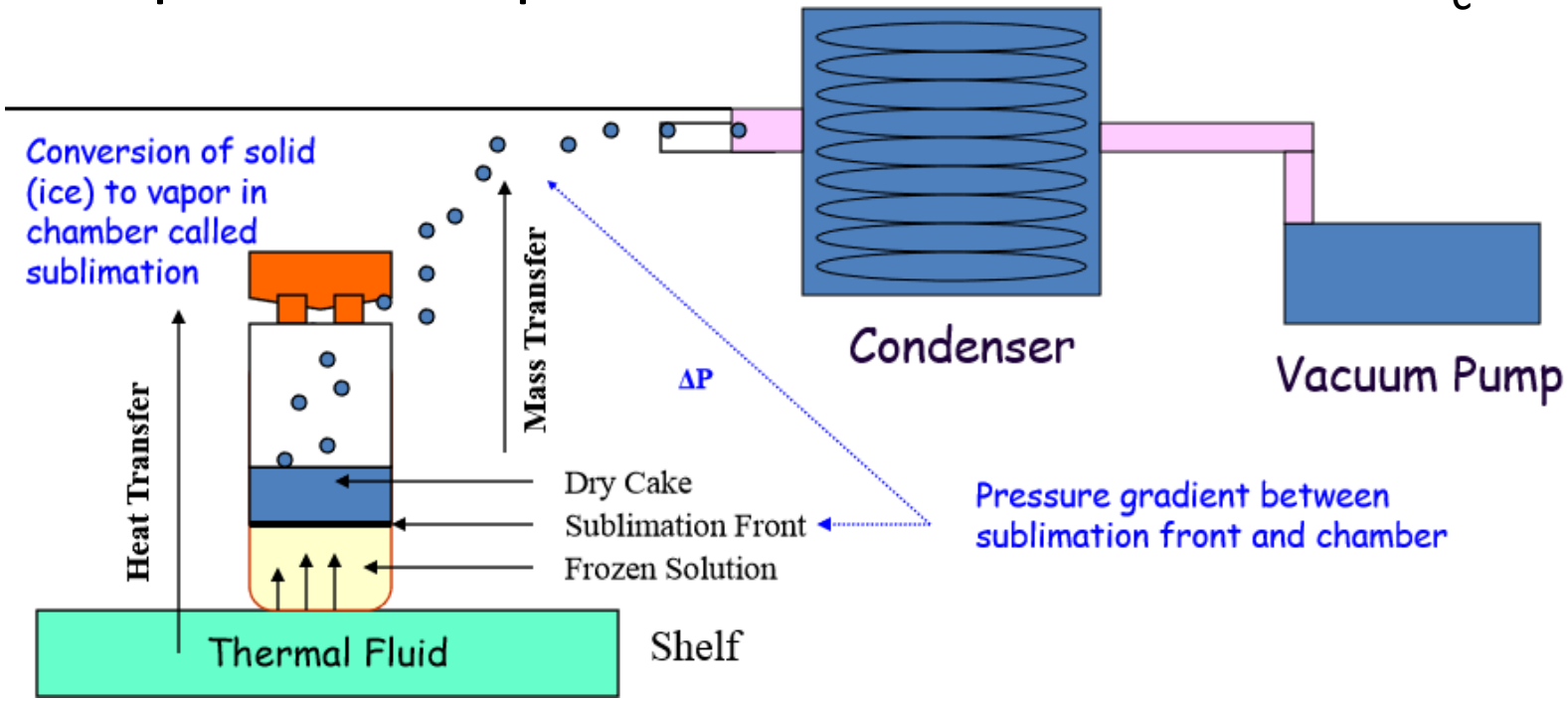
Must happen below collapse temperature, e.g. T_c = -31.4 C

Freeze-drying steps: freezing, annealing, primary drying, secondary drying

Biopharma Technology Ltd (BTL),n.d.

Primary Drying

- The sublimation of ice from the frozen product to achieve a dried layer of solute
- The product temperature should be lower than T_c



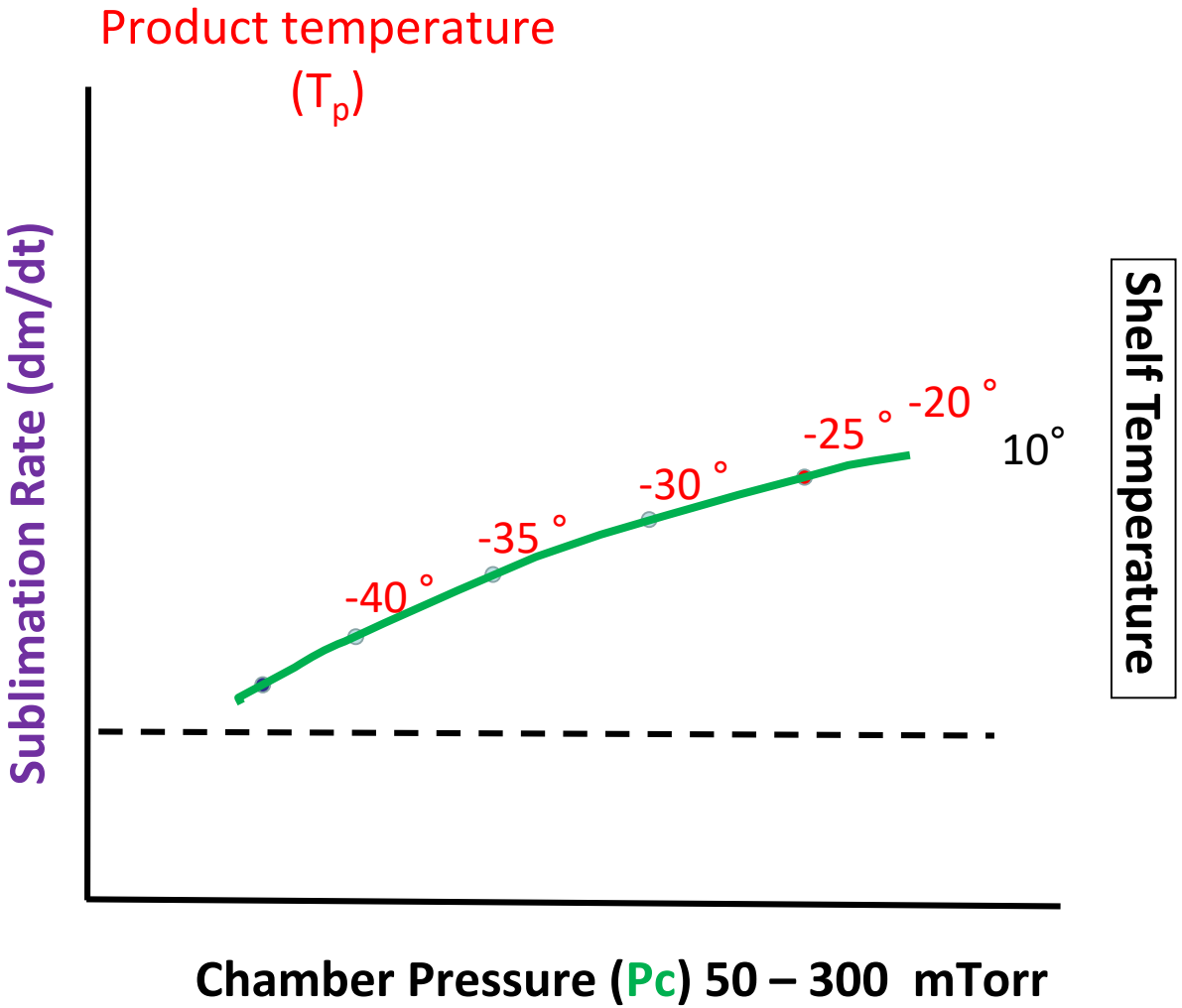
Heat and mass transfer in primary drying

Schwegman, 2011

Design Space for Primary Drying

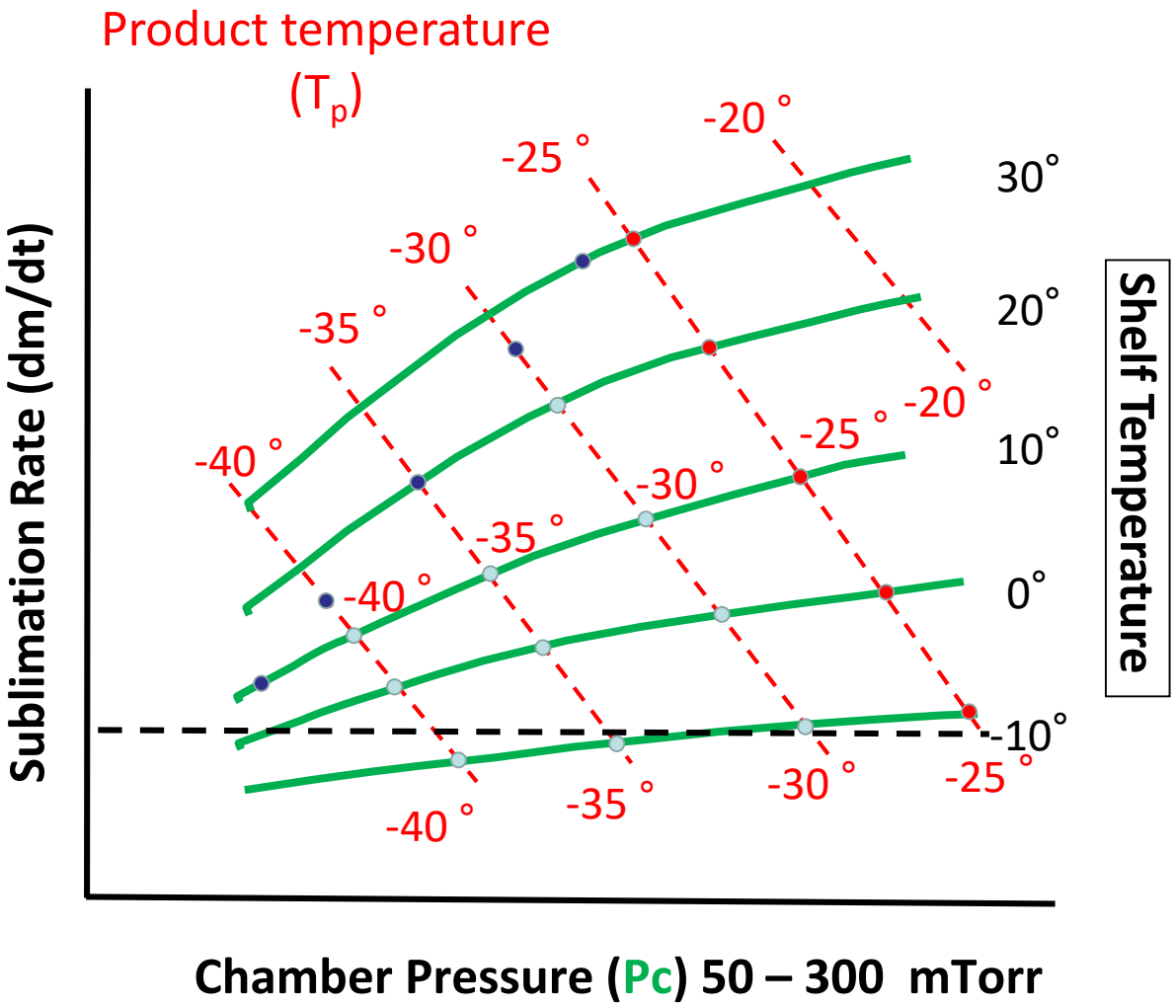
Design Space

Use a range of **chamber pressures** at a **fixed shelf temperature** (equipment settings) to establish the relationships between sublimation rate and the product temperature



Design Space

Repeat at different shelf temperatures

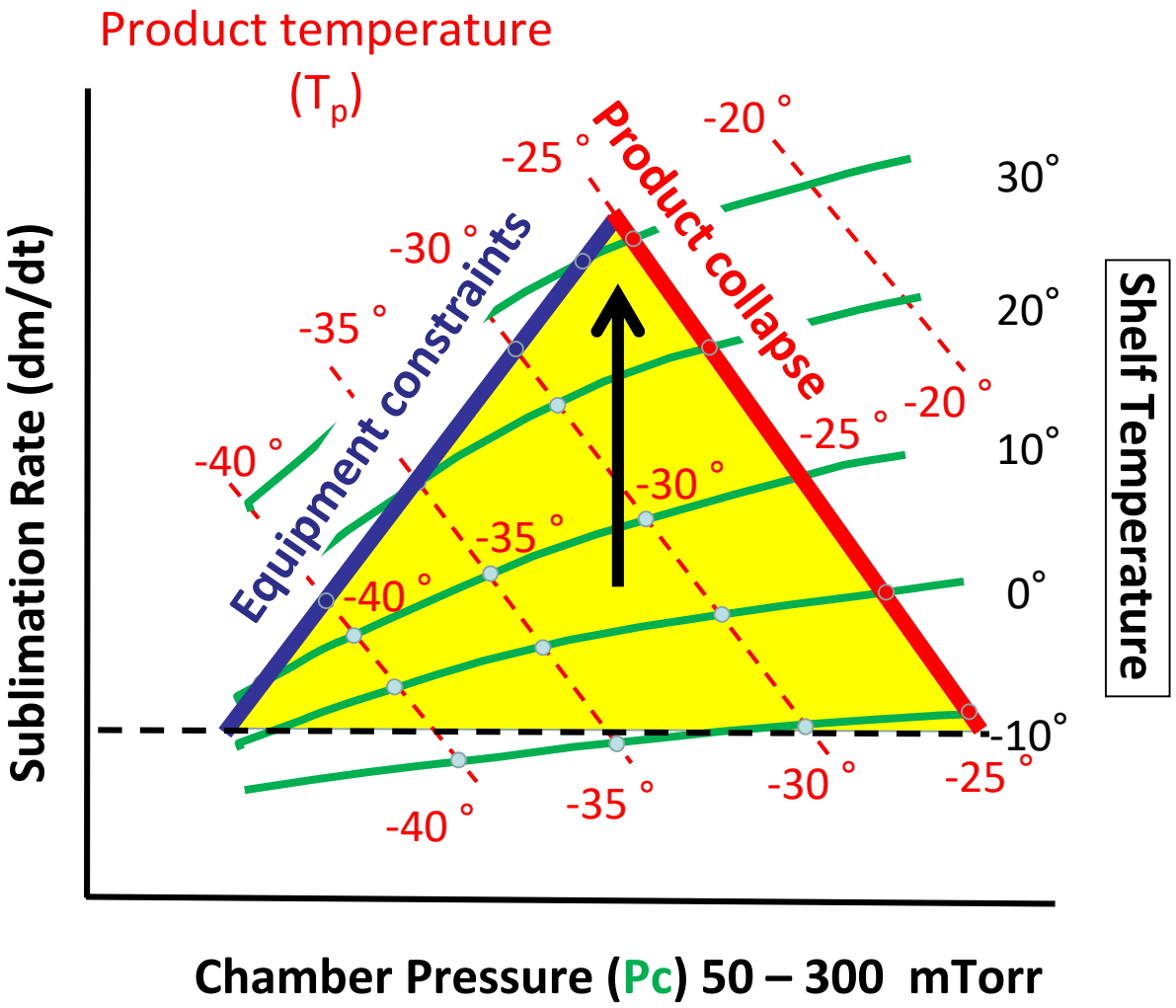


Design Space

And then map on the equipment constraints (chocked flow) ($P_i = P_c$) and the product constraint (collapse) $T_i = T_c$

To complete the design space (as shown by the yellow triangle)

Then operate at the apex of the triangle to drive process efficiencies



Through Vial Impedance Spectroscopy (TVIS)

Recent Funding

LyoDEA
£217 160 Collaborative R&D funding
(Nov ’08-Oct ’12) Innovate UK

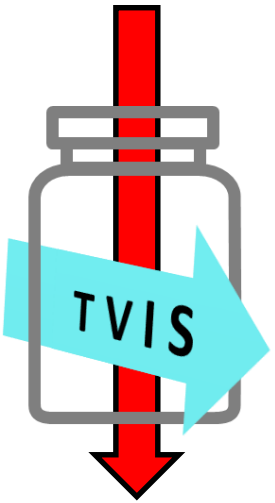
BioStaRT
£367 567 Collaborative R&D funding
(Aug ’14- Jul ’17) Innovate UK

AtlasBio
£803 846 Collaborative R&D funding
(Oct ’16-Sept 18) Innovate UK

IP: GB2480299
Electrical monitoring of a lyophilization process
Priority Date: 12th May 2010
Assignee: GEA Pharma Systems

Through Vial Impedance Spectroscopy (TVIS)

- Impedance measurements across a vial rather than within the vial
- Hence "**Through Vial Impedance Spectroscopy**"

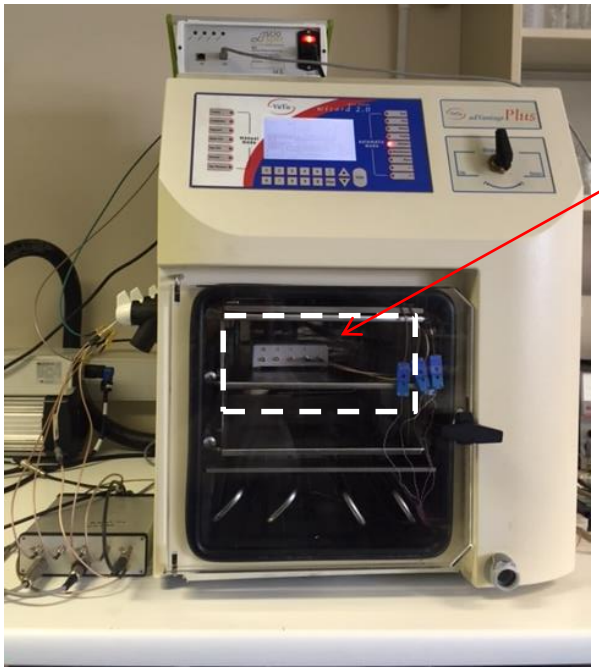


- Features
- Single vial "Non-product invasive"
- Both freezing and drying characterised in a single technique
- Stoppering still possible and on-perturbing to the packing of vials
- Mesoscale of unfrozen fraction accessible by assessing the temperature dependence of impedance

- On going TVIS development for multiple scales with top down electrodes:
  - Product Development (Microplate, Micro-vial, or Single vial)
  - Mini-Pilot (Small population clusters of vials)
  - Scale up to Batch (Large population cluster of vials)

Through Vial Impedance Spectroscopy (TVIS)

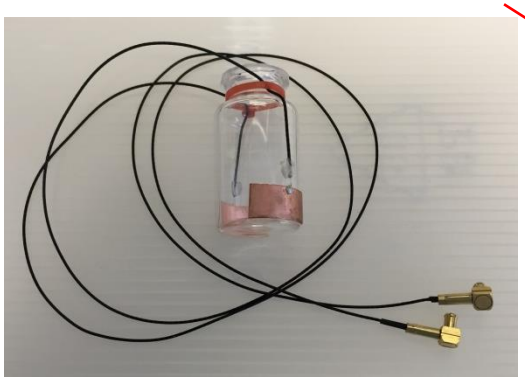
Freeze Dryer and TVIS System



Virtis Advantage Pro Freeze Dryer

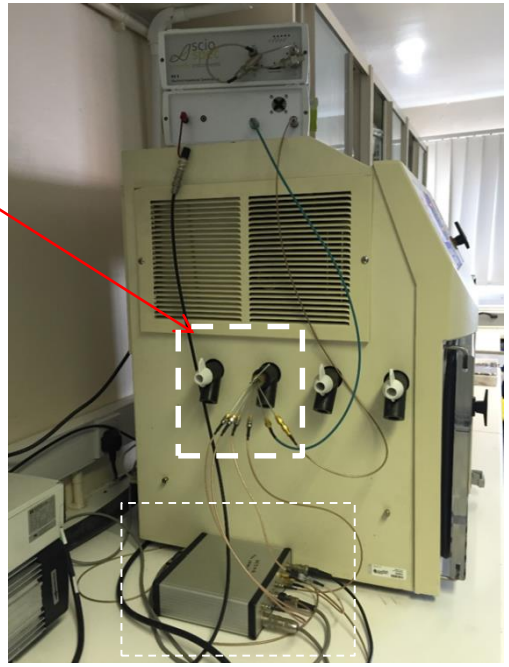
Junction box

Pass-through



Impedance test vial

TVIS System

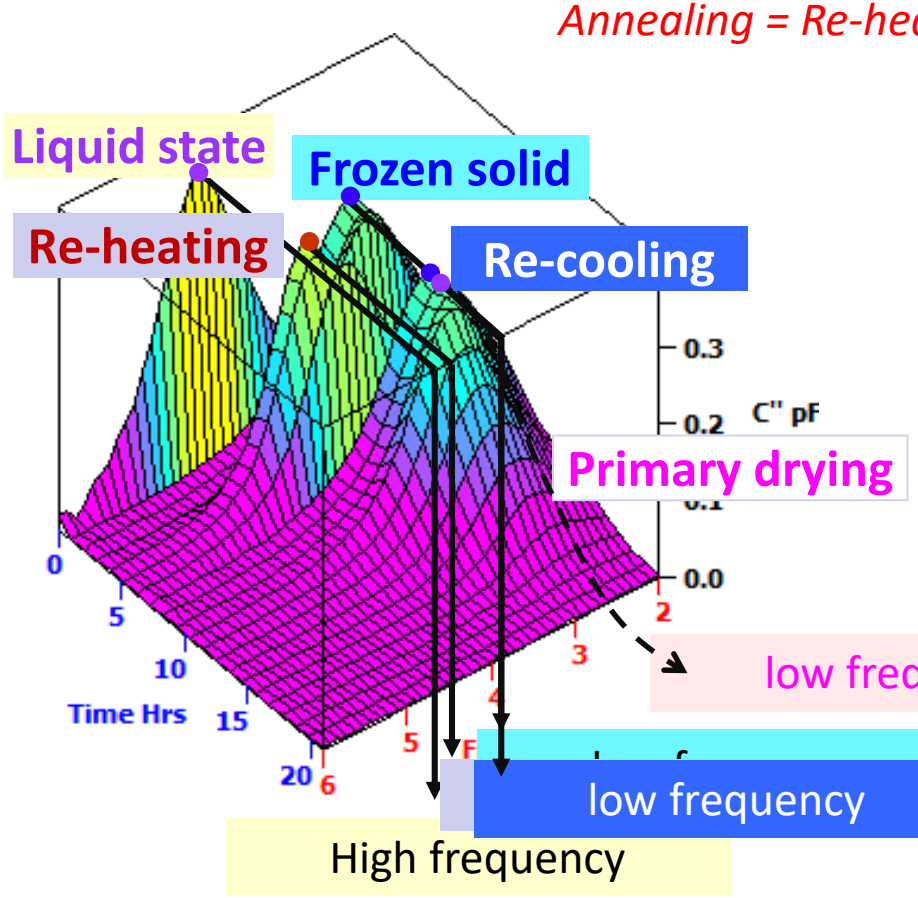


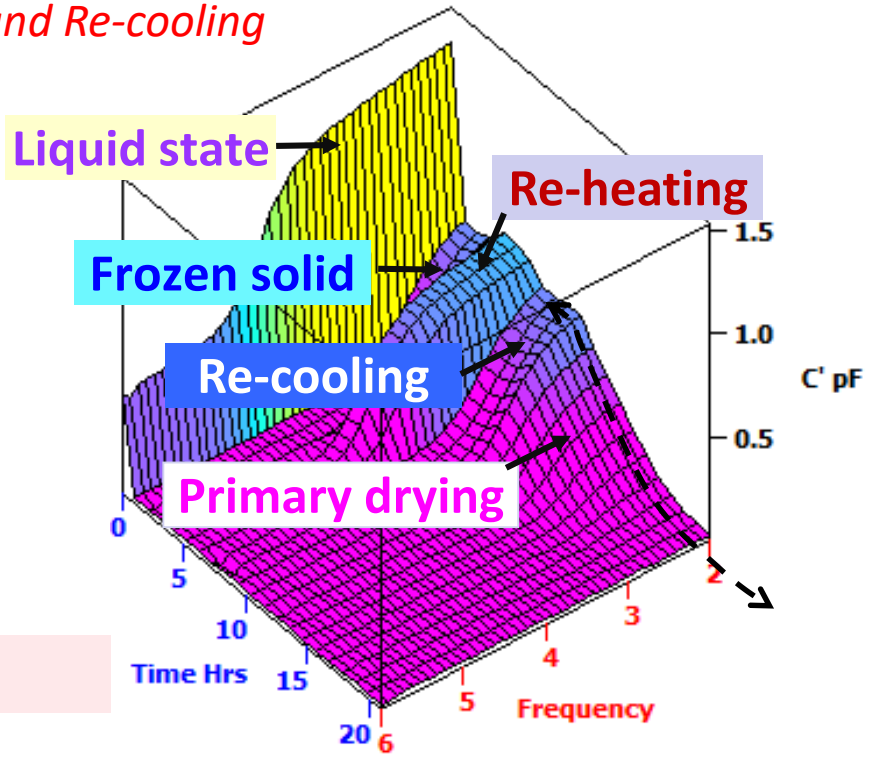
TVIS Response Surface

Imaginary Part of Capacitance

Real Part of Capacitance

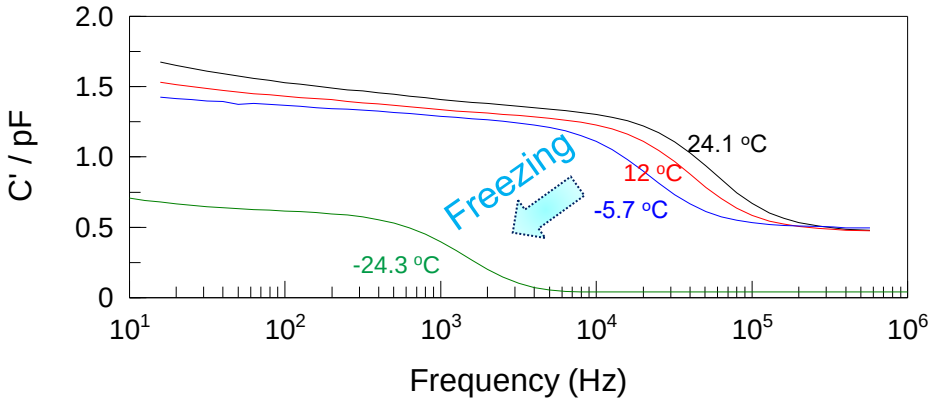
Annealing = Re-heating and Re-cooling

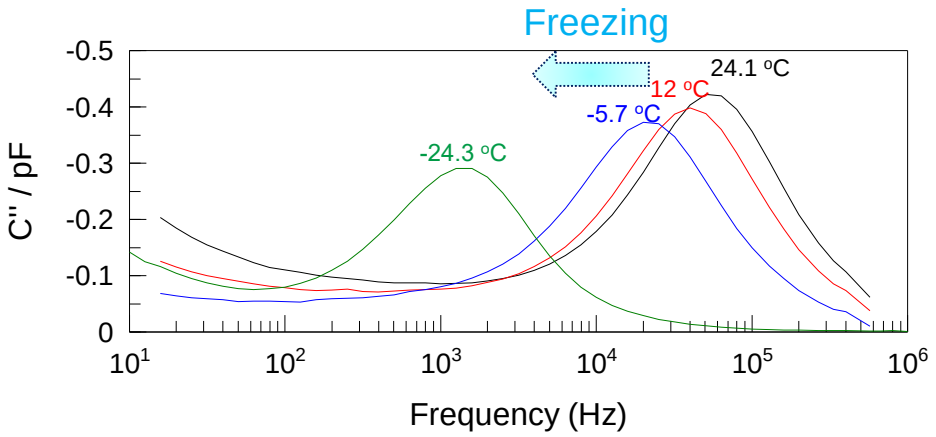




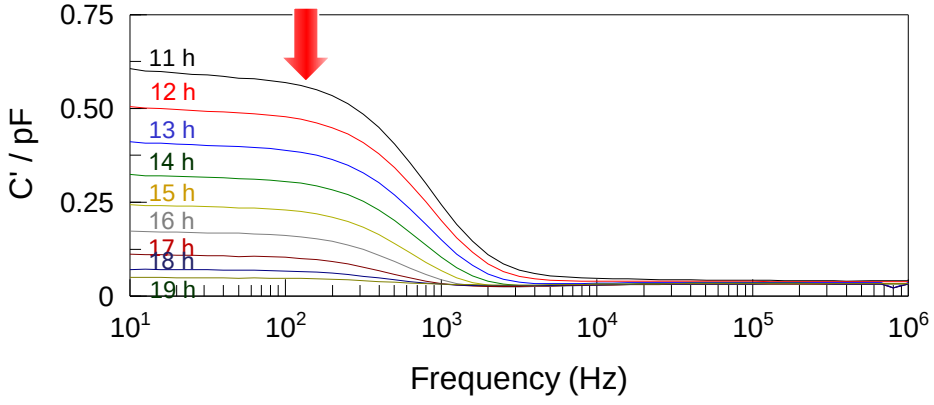
TVIS Response Surface

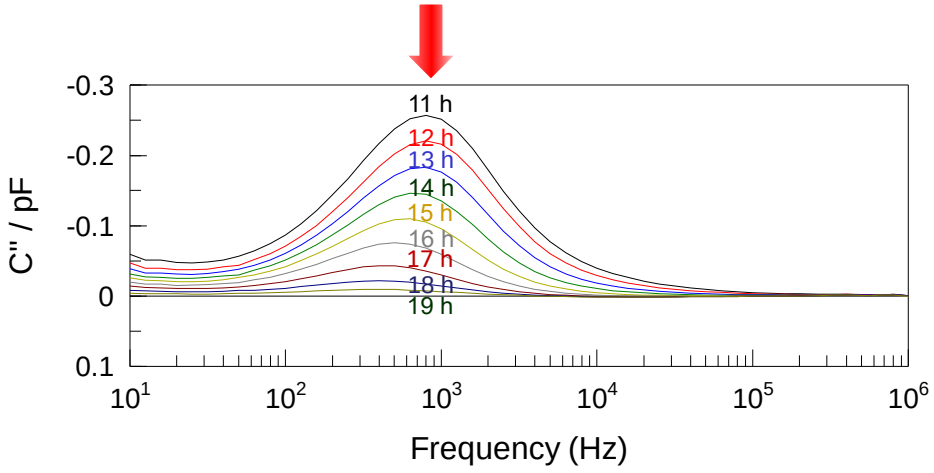
Freezing Process





Primary Drying Process





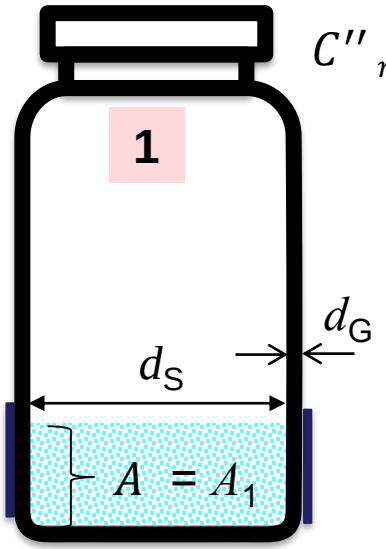
- The magnitudes of C_G and C_S are proportional to the area of interface between the frozen mass and the glass adjacent to the electrode (A).

$$C_G = \frac{\varepsilon_G \varepsilon_0}{d_G} A$$

Constant (K_G)

$$C_S = \frac{\varepsilon_S \varepsilon_0}{d_S} A$$

Constant (K_S)

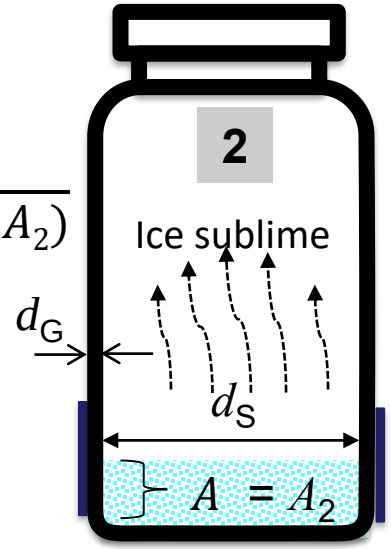


$$C''_{max1} = \frac{C_G^2}{2(Cs + CG)} = \frac{(K_G A_1)^2}{2(K_S A_1 + K_G A_1)} = kA_1$$

$$C''_{max2} = \frac{C_G^2}{2(Cs + CG)} = \frac{(K_G A_2)^2}{2(K_S A_2 + K_G A_2)} = kA_2$$

Primary Drying

$$C''_{max1} > C''_{max2}$$



- In case of a flat sublimation interface, the interfacial area between the frozen layer and the juxtaposed glass wall (A) will decrease in proportion to the remaining ice volume

TVIS Technology: Principle

- The capacitance spectrum is dependent on both *the electrical resistance and electrical capacitance of the vial contents.*
  - Data viewing software (LyoView ™) identifies the peak frequency (F_{PEAK}) and peak amplitude (C''_{PEAK}) in the imaginary part of the capacitance spectrum
- In general terms:
  - F_{PEAK} can be used to monitor phase behaviour (ice formation, glass transitions) and product temperature
  - C''_{PEAK} can be used to monitor the amount of ice remaining during primary drying, from which the drying rate and end point may be determined.

TVIS Application
Heat Transfer Coefficient (KV) Determination
(Test sample: 2 mL water in 10 mL vial)

Primary Drying Modelling : Heat and Mass Transfer

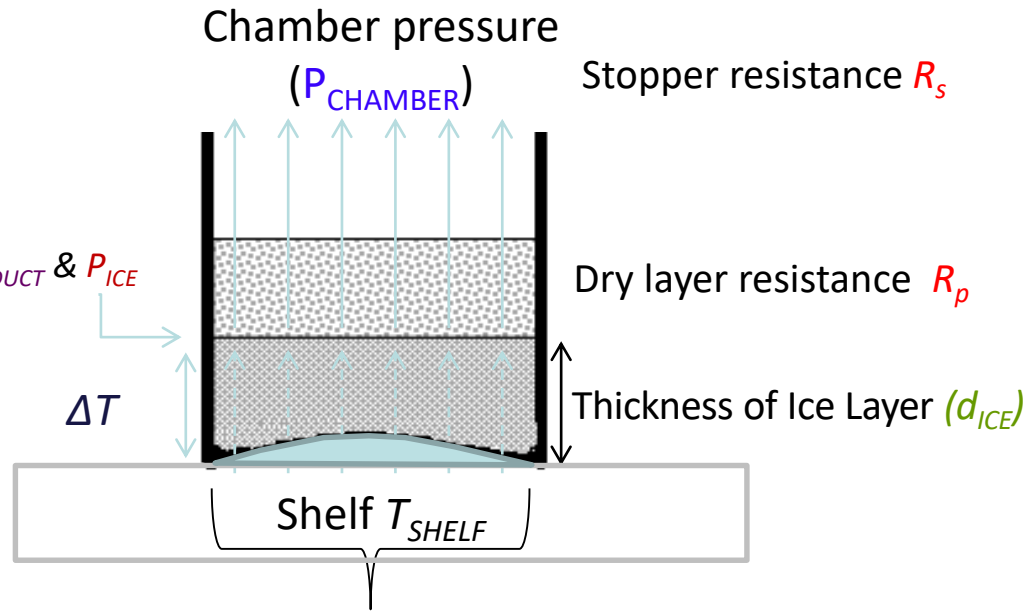
$$\frac{dm}{dt} = \left(\frac{P_{ICE} - P_{CHAMBER}}{\hat{R}_{PS}}\right) \cdot A_P$$

$$\frac{dq}{dt} = L\left(\frac{dm}{dt}\right)$$

K_V determination

$$\frac{dq}{dt} = A_V \boldsymbol{K_V} (T_{SHELF} - T_{PRODUCT})$$

$$\ln(PICE) = \left(\frac{-6144.96}{T_{PRODUCT}}\right) + 24.02$$



dm/dt - sublimation rate (g/hour/vial)

P_{ICE} - Vapour pressure at ice sublimation interface (Torr)

$P_{CHAMBER}$ - Chamber pressure (Torr)

$\hat{R}_{PS}$ - Area normalized product and stopper resistance

dq/dt - heat flow to the product

L - Latent heat of sublimation (J g^{-1})

A_V - cross-sectional area of the vial (cm^2)

T_{SHELF} - Shelf temperature (K)

$T_{PRODUCT}$ - Product temperature at the bottom of vial (K)

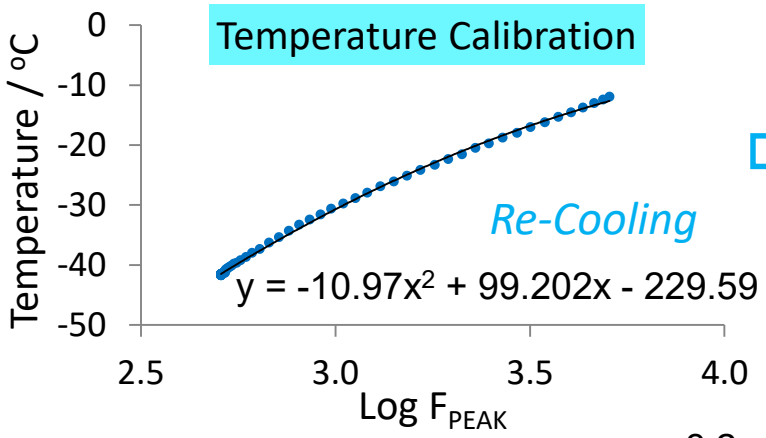
d_{ICE} - Thickness of the ice layer calculated (cm)

ΔT - Temperature different between ice sublimation front and the bottom of the vial (K)

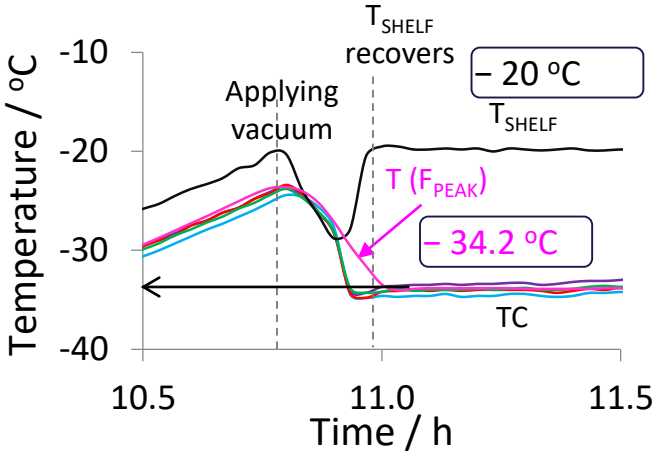
TANG, X.C., NAIL, S.L. and PIKAL, M.J. (2005) Freeze-drying process design by manometric temperature measurement: design of a smart freeze-dryer. Pharmaceutical Research, 22 (4), pp. 685-700.

Heat Transfer Coefficient (KV) Determination

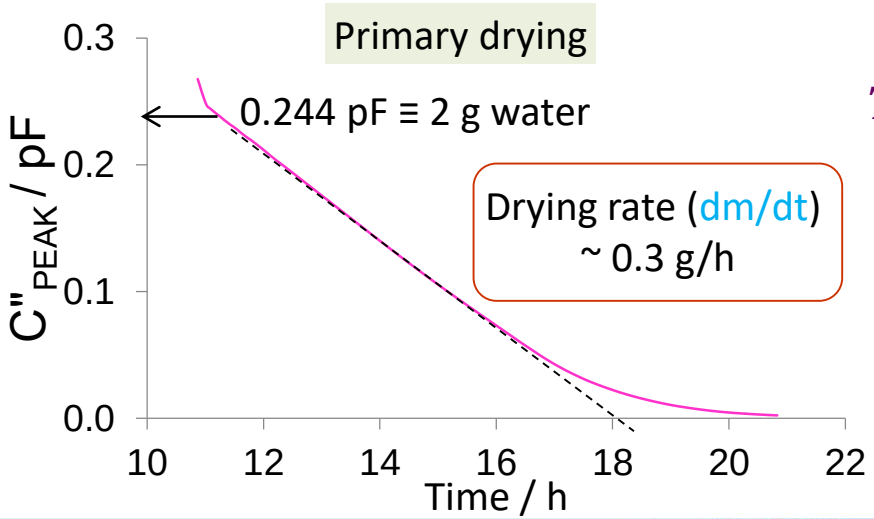
- The product temperature ($T_{PRODUCT}$), which derived by ***TVIS*** as $T(F_{PEAK})$ or *by thermocouple (TC),* is one of parameters needed for K_V determination



Calculate $T_{PRODUCT}$ from Log F_{PEAK} (TVIS)



- Sublimation rate (*dm/dt*) is estimated by ***TVIS***
- Test sample is 2 mL water filled in 10 mL vial



$T_{PRODUCT}$ and *dm/dt* from ***TVIS***

K_V determination

Heat Transfer Coefficient (KV) Determination

- First convert dm/dt to dq/dt using the latent heat of sublimation
  (L = 2844 J g^{-1})

$$L \frac{dm}{dt} = \frac{dq}{dt}$$

dm/dt = 0.3 g/h

L = 2844 J g^{-1}

dq/dt = 853 J/h

$$\frac{dq}{dt} = A_V K_V (T_{SHELF} - TPRODUCT)$$

T_{SHELF} = – 20 °C

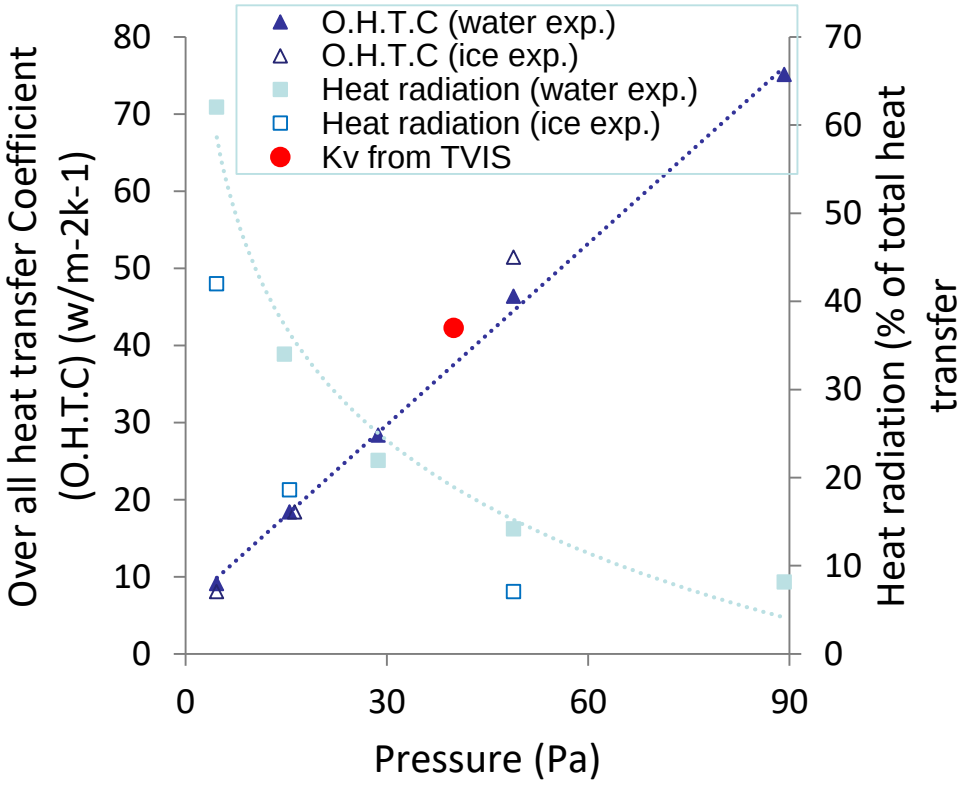
$T_{PRODUCT}$ = – 34.2 °C

A_v = 0.0045 m^2

(*Schott Type1 glass 10 ml tubing vial*)

K_v = 37 W m^{-2} K^{-1} [@ 40 Pa, 400 µBar]

K_v values for 10 mL tubing vials (2 mL fill volume)

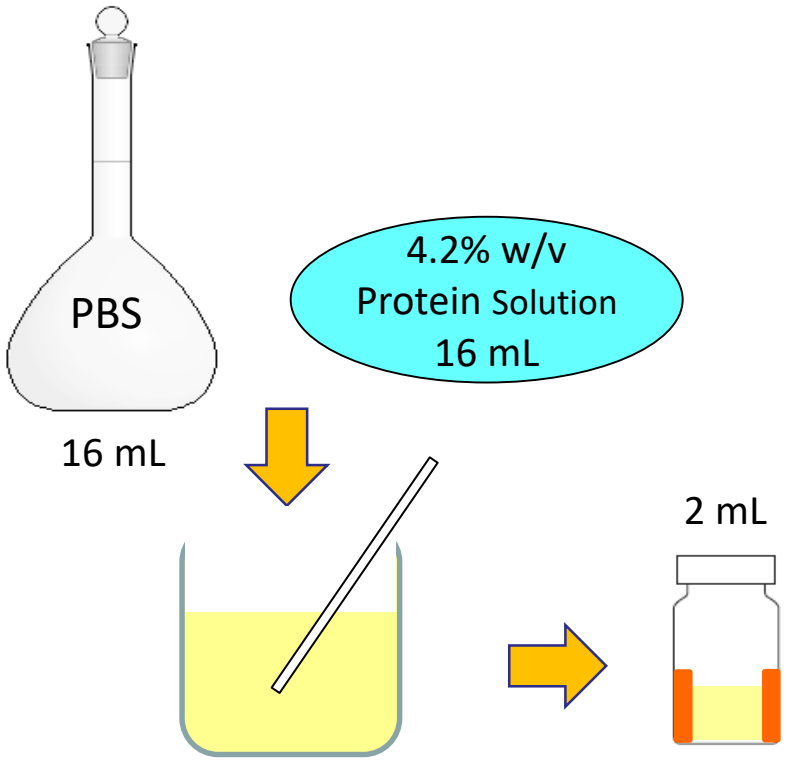


Brülls, M., and Ramusson, A. (2002) Heat Transfer in Lyophilization. Int J Pharm 10;246(1-2):1-16.

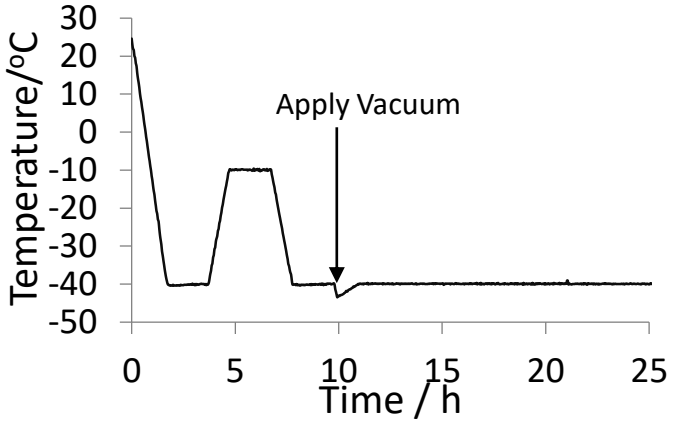
TVIS Application
Product Resistance (Rp) Determination
(Test sample: 2 mL protein solution fill in 10 mL vial)

Dried Product Resistance (RP) Determination of Protein Solution

Preparation: 50% v/v Protein Solution



Freeze drying Protocol

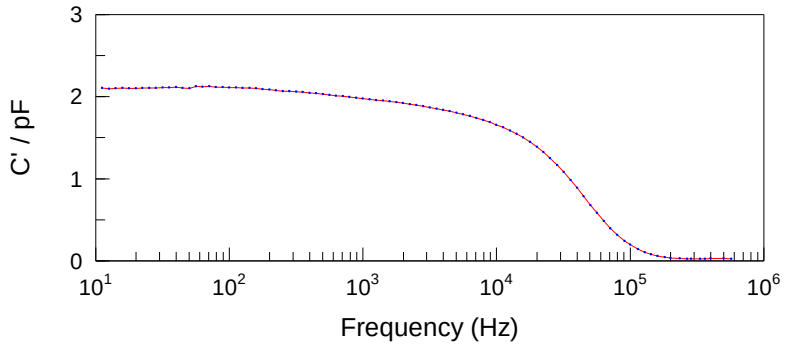


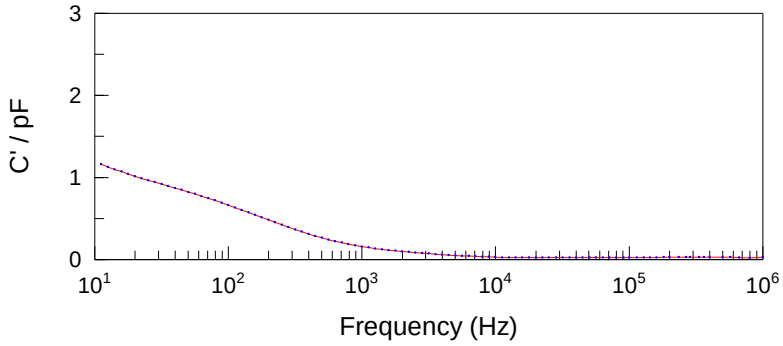
- A 50% v/v protein solution 2 mL has protein 0.042 g
- Protein 1 g has ~0.3 g (unfrozen water) unavailable water (Pang 2014)
- Therefore unfrozen water of protein is 0.3 x 0.042 = 0.0126 g
- The weight of ice is approximately 2 – 0.042 - 0.0126 = 1.932 g

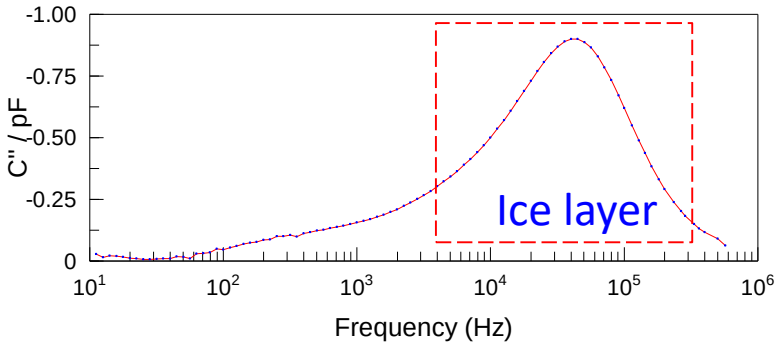
Pang, X.-F. (2014) *Water: Molecular structure and properties*. Singapore, Singapore: World Scientific Publishing Co Pte.

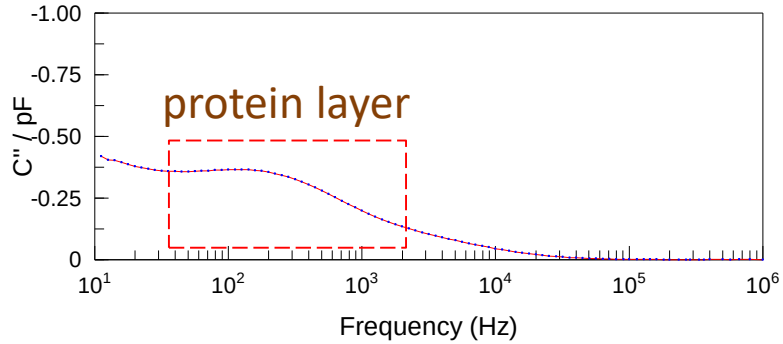
Product characterization: Opportunity for Protein Solution

- Low frequency process may hold information on the state/properties of the protein layer, inc. progression and end point of secondary drying





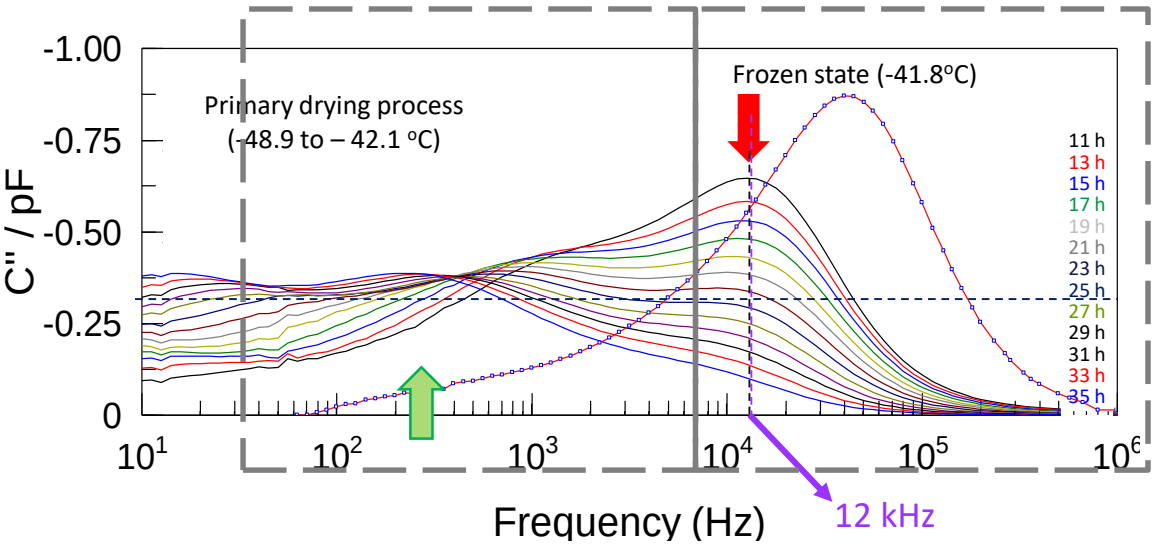


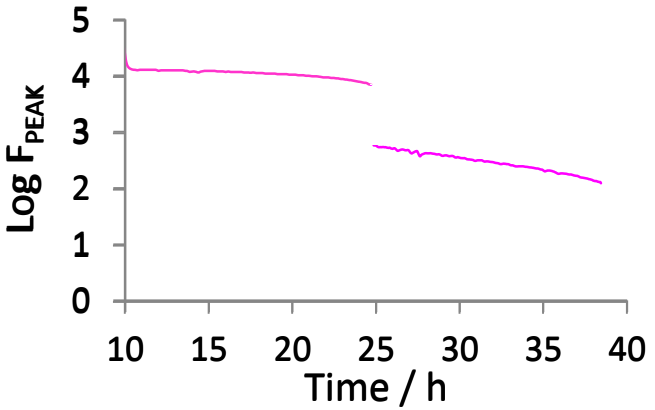


High frequency process (start of primary drying)

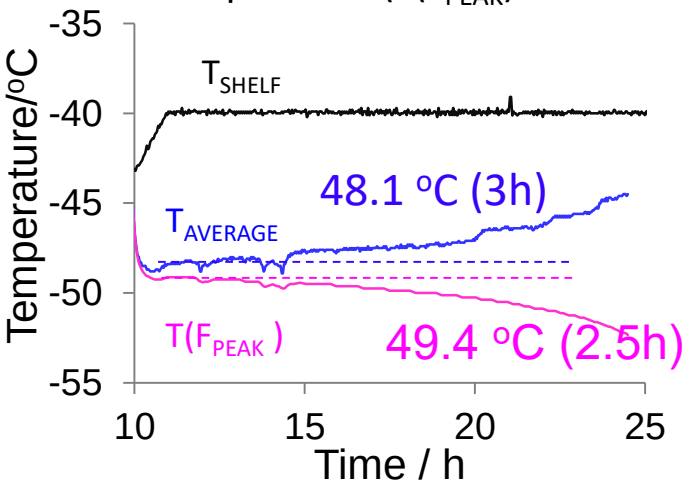
Low frequency process (end of primary drying)

Dried Product Resistance (RP) Determination of Protein Solution





Calibration data for log F_{PEAK} during annealing is used to predict the product temperature ($T(F_{PEAK})$)



- The peak at high frequency (~12 kHz) is due to the ice layer
  - Ice layer peak **decreases** during the early stages of primary drying (< 25 h) because the peak amplitude is proportional to the volume of the ice layer
- The peak at low frequency is due to the dry layer
  - Dry layer peak **increases** during the later stages of primary drying (> 27h) because the peak amplitude is proportional to the volume of the dry layer

Dried Product Resistance (RP) Determination of Protein Solution

- The different types of resistance to mass transport (water vapour flow)

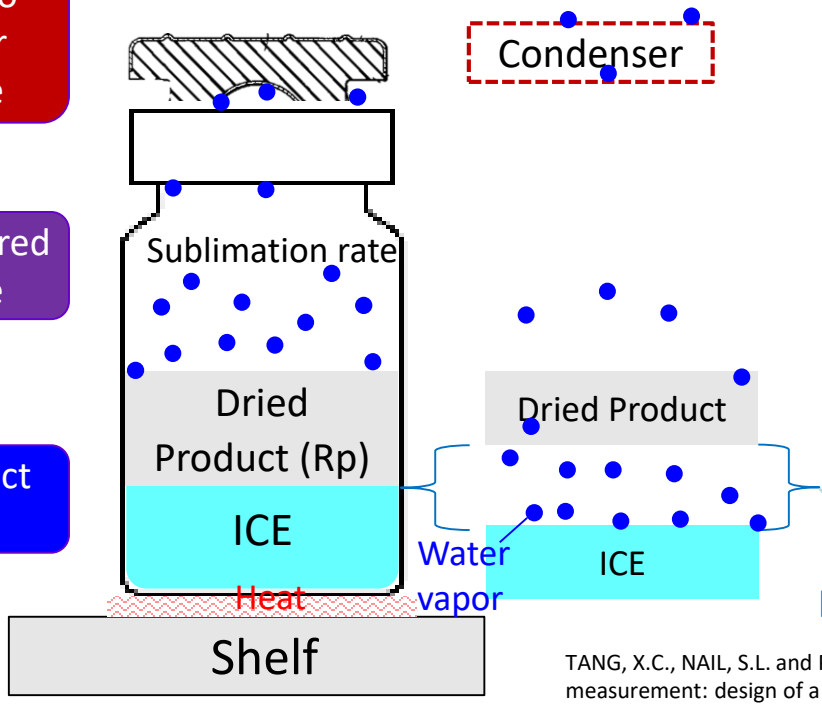
$$\frac{dm}{dt} = \left(\frac{P_{ICE} - PCHAMBER}{R_P + RS + RC}\right) \Rightarrow \frac{dm}{dt} = \left(\frac{P_{ICE} - P_{CHAMBER}}{\hat{R}p}\right) \cdot A_P$$

Product resistance (R_P) is ~ 80% of the total mass transfer resistance (Pikal, Roy and Shah (1984).

Chamber to condenser resistance

Semi-stoppered resistance

Dried product resistance



dm/dt = the rate of mass transfer for the water vapour or sublimation rate (g/hour/vial)

P_{ICE} = the equilibrium vapour pressure of ice at the sublimation interface temperature (Torr)

$P_{CHAMBER}$ = the chamber pressure (Torr)

$\hat{R}_p$ = the area normalized resistance of the dried product ($cm^2 \cdot Torr \cdot h \cdot g^{-1}$)

A_p = the cross-sectional area of the product (cm^2)

TANG, X.C., NAIL, S.L. and PIKAL, M.J. (2005) Freeze-drying process design by manometric temperature measurement: design of a smart freeze-dryer. Pharmaceutical Research, 22 (4), pp. 685-700.

Dried Product Resistance (RP) Determination of Protein Solution

$$\frac{dm}{dt} = \left(\frac{P_{ICE} - PCHAMBER}{\hat{R}p}\right) \cdot A_P$$

Ice vapour pressure calculated from $T_{PRODUCT}$ the product temperature (as derived by ***TVIS*** *or Thermocouple*)

Chamber pressure calculated from the temperature of condenser ($T_{CONDENSER}$) (in case of condenser is in the freeze dry chamber)

$$\hat{R}p = \left(\frac{P_{ICE} - PCHAMBER}{\frac{dm}{dt}}\right) \cdot A_P$$

$$\ln(P_{ICE}) = \left(\frac{-6144.96}{T_{PRODUCT}}\right) + 24.02$$

$$\ln(PCHAMBER) = \left(\frac{-6144.96}{T_{CONDENSER}}\right) + 24.02$$

dm/dt can estimate by **TVIS**

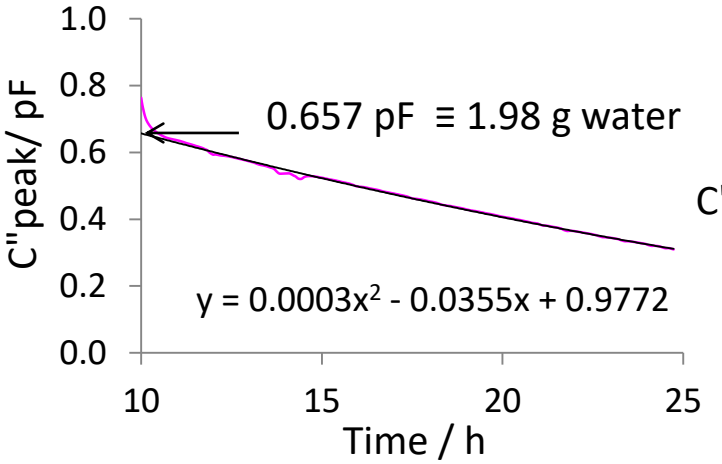
Cross-sectional area of product is calculated by using internal diameter of test vial

For example, Schott Type1 glass 10 ml tubing vial has internal diameter 2.2 cm therefore,

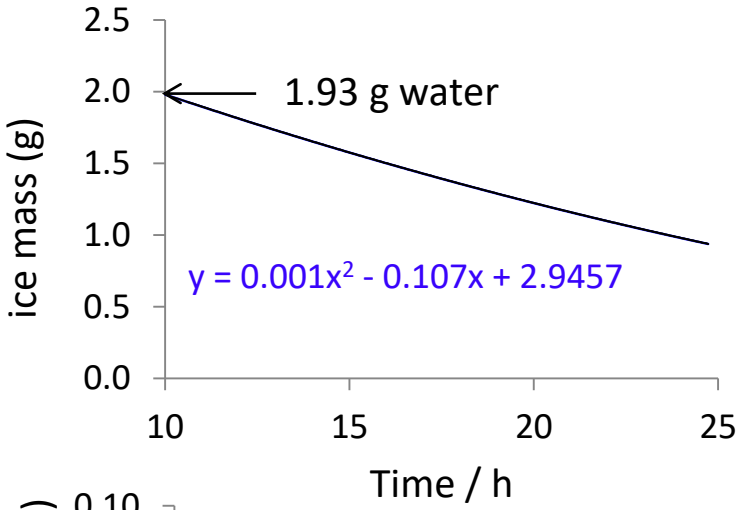
A_P = 3.8 cm²

Dried Product Resistance (RP) Determination of Protein Solution

- The drying rate (*dm/dt*) estimated by ***TVIS*** is one of parameters used for determination of dried product resistance (R_p)
- First convert C''_{PEAK} from TVIS to Ice mass



C''_{PEAK} is proportional to the amount of ice



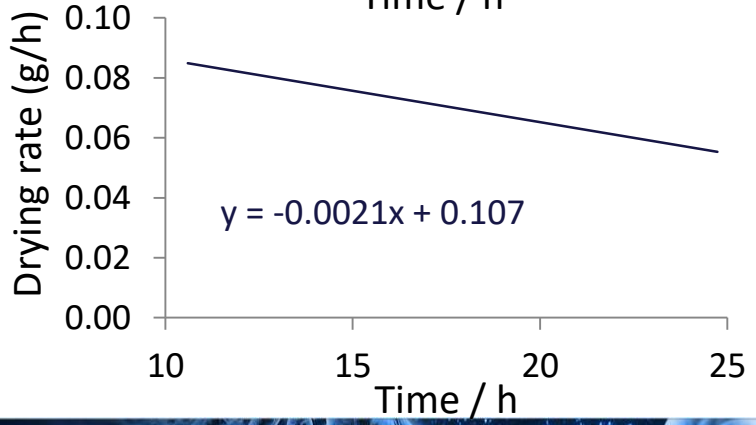
- Then, calculate drying rate (*dm/dt*) using time derivative of ice mass

$y = 0.001x^2 - 0.107x + 2.9457$

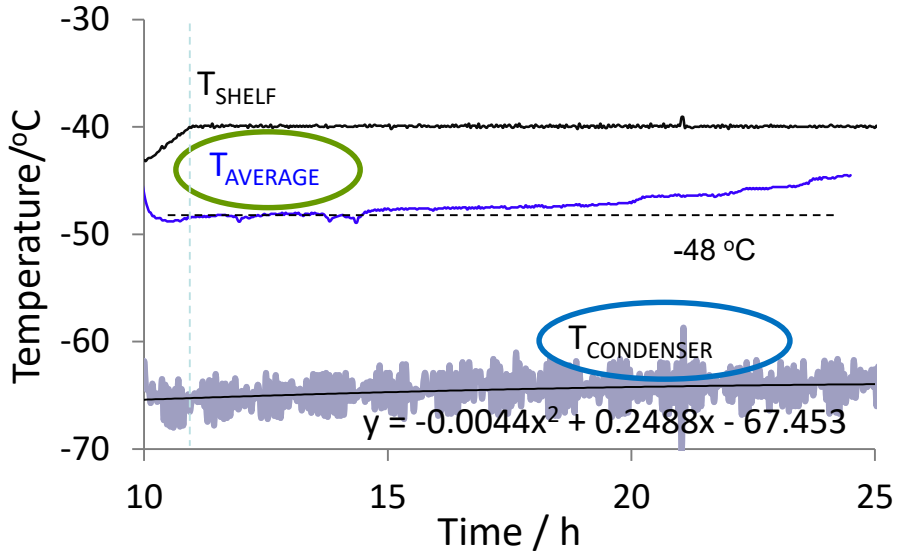
↓

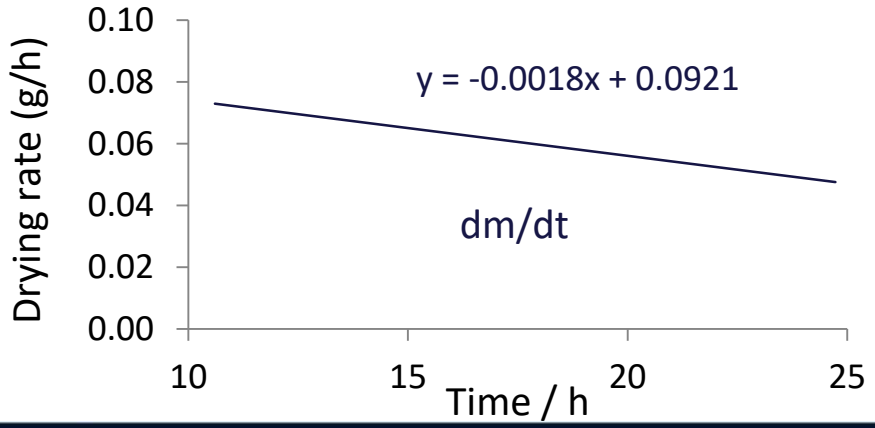
$dm/dt = 0.002X - 0.107$

R_p determination



Dried Product Resistance (RP) Determination of Protein Solution





$$\ln(P_{ICE}) = \left(\frac{-6144.96}{T_{PRODUCT}}\right) + 24.02$$

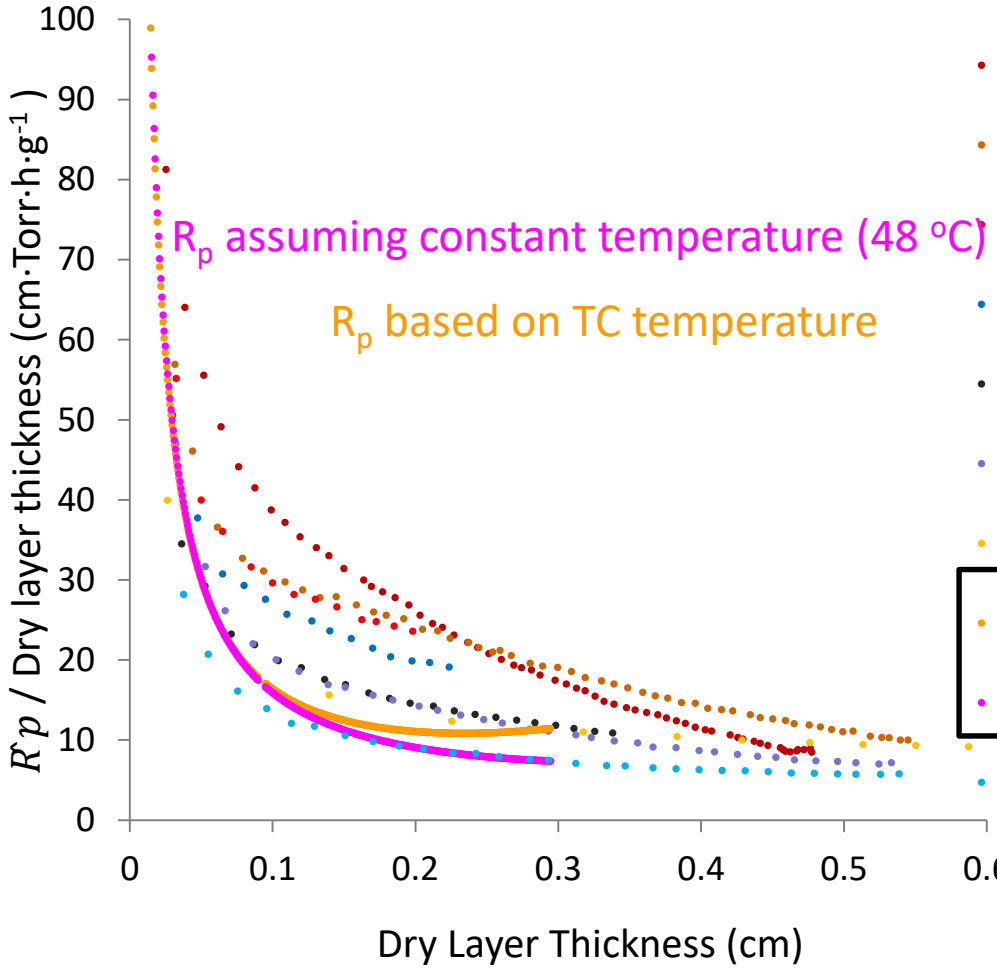
$$\ln(P_{CONDENSER}) = \left(\frac{-6144.96}{T_{CONDENSER}}\right) + 24.02$$

A_p = 3.8 cm²
(Schott Type1 glass 10 ml tubing vial)

dm/dt

$$\hat{R}p = \left(\frac{P_{ice} - PC}{\frac{dm}{dt}}\right) \cdot A_P$$

Dried Product Resistance (RP) Determination of Protein Solution



- 5% BSA annealing 1 h (Ref. 3)
- 2% BSA annealing 1 h (Ref. 3)
- 2% BSA non annealing (Ref. 4)
- 2% IgG non annealing (Ref. 4)
- 2% Lysozyme non annealing (Ref. 4)
- 0.5% BSA annealing 6 h (Ref. 3)
- 5% Glycine non annealing (Ref. 2)
- 4% Protein solution annealing 1 h (poly 3rd)
- 4% Protein solution annealing 1 h (constant)
- 0.5% BSA annealing 1 h (Ref. 3)

1. GIESELER, H., KRAMER, T. and PIKAL, M.J. (2007); 2. JOHNSON, R.E. et al. (2010); 3. LEWIS, L.M. et al (2010)

Summary & Future Work

- K_v value determination possible.
- Increase the drying time over which T(Feak) maybe determined by increasing the fill volume of ice.
- Investigate the dependence of K_v son chamber pressure for design space determination

- R_p value determination possible:
- Investigate uncertainties in temperature measurement to provide for more reliable estimates of Rp (compare RTDs vs TCs)
- Calculate Product Temperature for heat flux (dq/dt) and Kv
- Use equivalent circuit modelling to fit spectra and develop a TVIS surrogate temperature model
- Characterise a wide range of materials, formulations and process parameters, inc. nucleation temperature, fill volume, freezing rate, annealing

Summary of Applications of TVIS

- TVIS registers thermal events through changes in the sample resistance associated with
  - Discontinuous changes in viscosity (glass transition, collapse)
  - Change of state (e.g. ice formation and eutectic formation)
- Temperature control might be possible through improved modelling and calibration (Equivalent circuits)
- Primary drying (loss of ice) is monitored through changes in the strength of the dielectric loss peak (or step in the real part capacitance)
  - Enables drying rate determinations for Kv and Rp calculations
- Meso-structural information extracted through the (non-Arrhenius) temperature dependence of the resistance
- Mechanisms of annealing may be elucidated from changes in resistance with time (during the heating-hold phase) and from the absence of any changes in T_G
- Future (with non-contact system)
  - Opportunities to track the physical characteristics over a range of scales, from micro-titre plates to collections of vials

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BIOPHARMACEUTICAL QUALITY BY DESIGN


Any
Questions?

TVIS Application
Other Applications

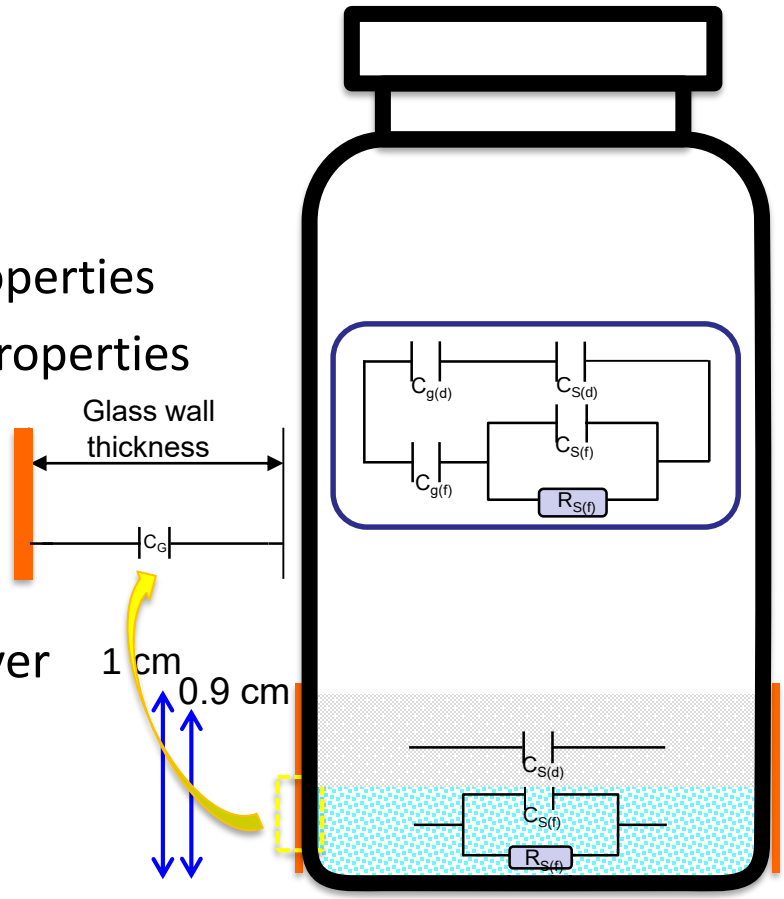
Through Vial Impedance Spectroscopy (TVIS)

- **Electrodes** are attached to the external surface of a vial
- The vial is filled with the sample to fill factor (Φ) of 0.9

$$\text{Fill factor } (\Phi) = \frac{\textit{The height of liquid fill}}{\textit{The height of active electrode}}$$

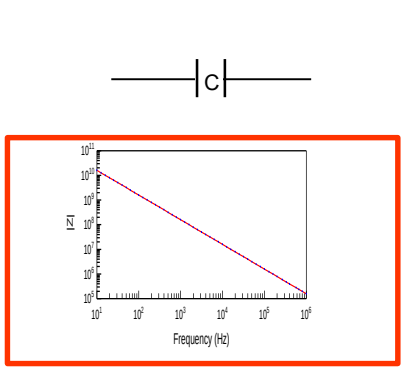
- The sample has both resistive & capacitive properties
- The glass vial has both resistive & capacitive properties
- But glass resistance is so high therefore it behaves primarily as a capacitor.
- In freeze drying, the sample is frozen and dried by sublimation to obtain a 'dry' layer
- The dry layer is predominantly capacitive due to its low moisture content / high resistance

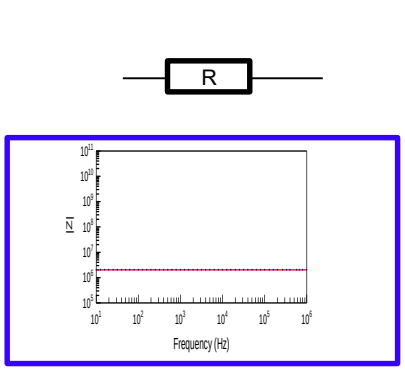
  Exception - Proteins

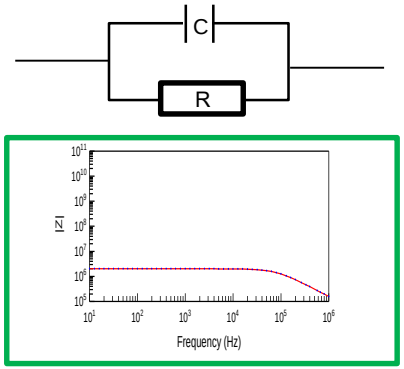


Through Vial Impedance Spectroscopy (TVIS)

- Impedance is a frequency dependent parameter largely because the impedance of a capacitance is dependent on the frequency of the applied field, whereas an ideal resistor has zero frequency dependence







- By fitting the impedance spectrum of a composite object then one can extract the sample resistance and capacitance

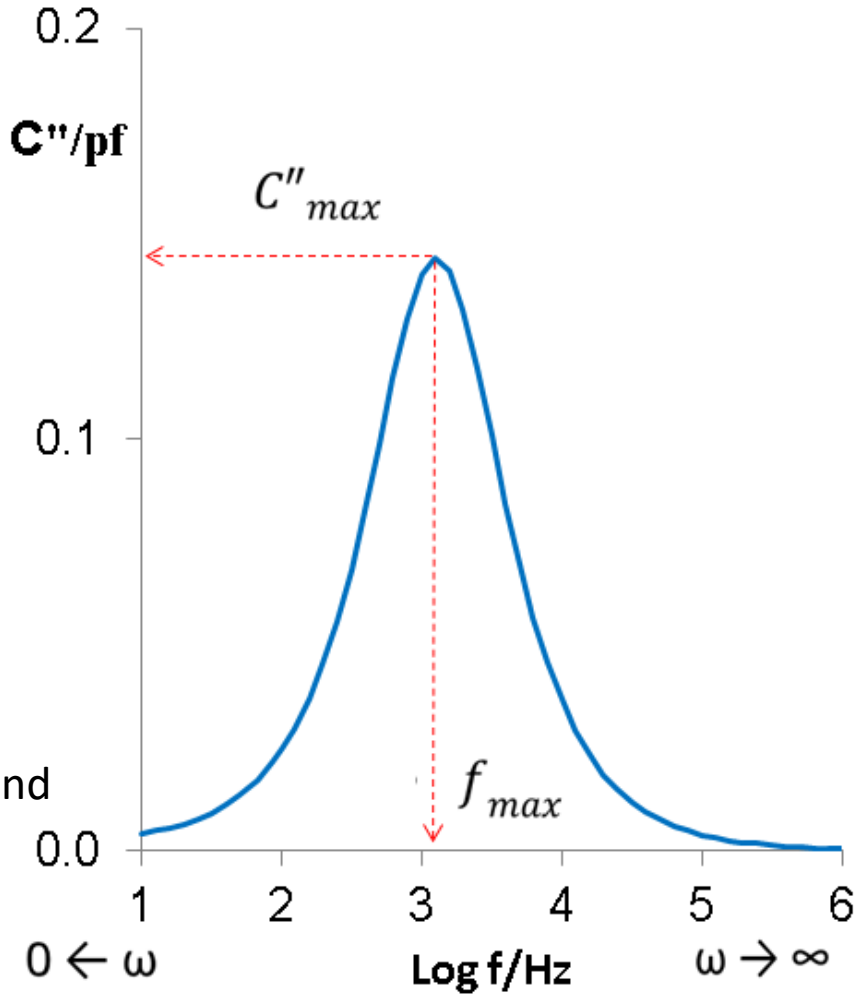
Imaginary Part Capacitance : Characteristic Response

- At ω→0, $C^{"} = 0$
- As the frequency increase, $C^{"}$ increases to maximum ($C^{"}_{max}$) then decreases to 0 as the frequency ω→∞
- At a frequency of

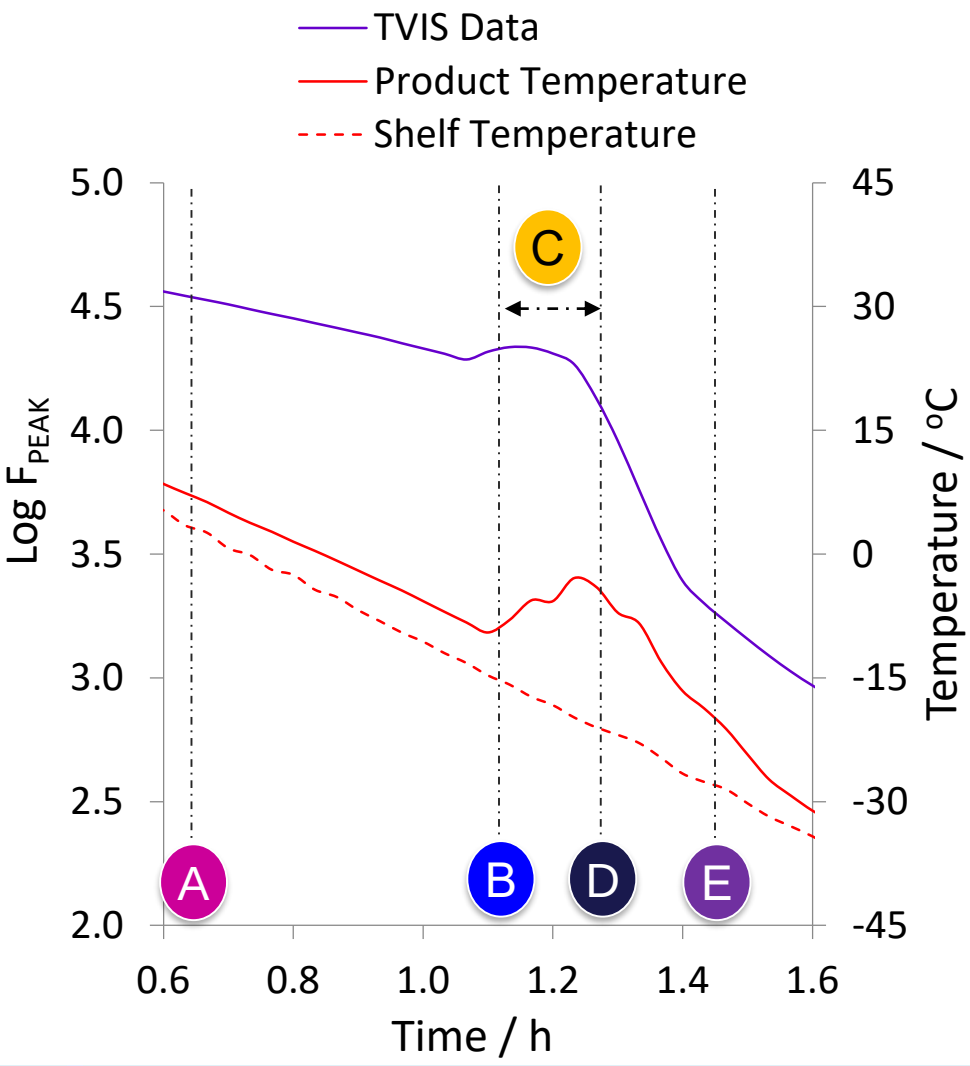
$$\omega_{max} = \frac{1}{R_S(C_s + C_G)} \text{ in radians}$$

$$f_{max} = \frac{1}{2\pi Rs(Cs+CG)} \text{ in cycles per second}$$

$$C''_{max} = \frac{C_G{}^2}{2(CS + CG)}$$



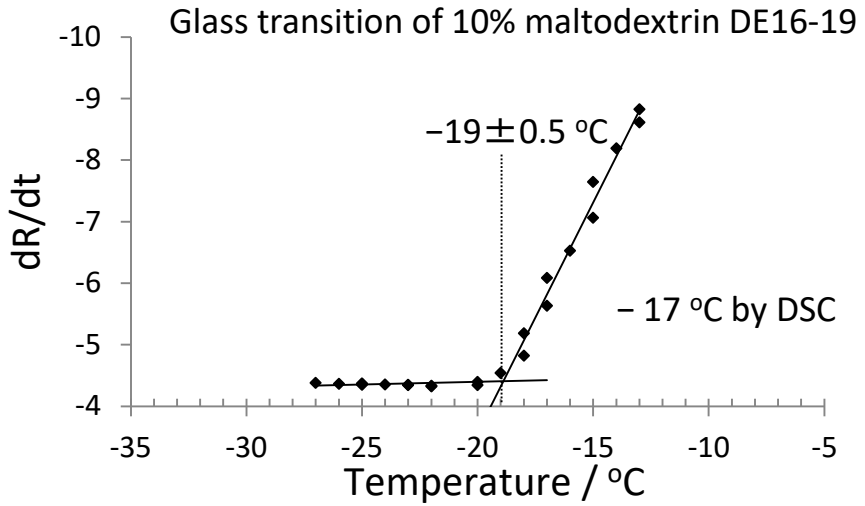
Product characterization



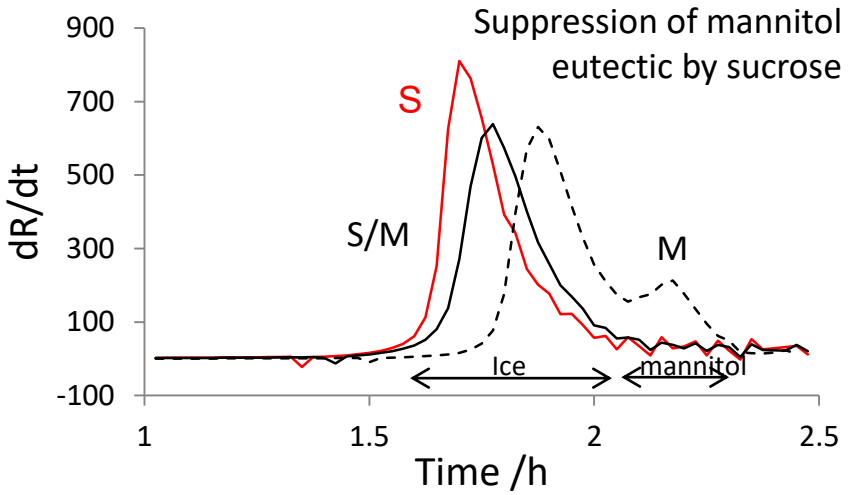
- Log F_{PEAK} profile of freezing step shows the ice nucleation process:
  - Solution is cooled at A
  - Solution super-cools and starts ice nucleation at B This points is referred as "*onset of ice nucleation*"
  - Crystallization or *ice nucleation formation* occurs during C period and continues until the *end point of solidification* at D

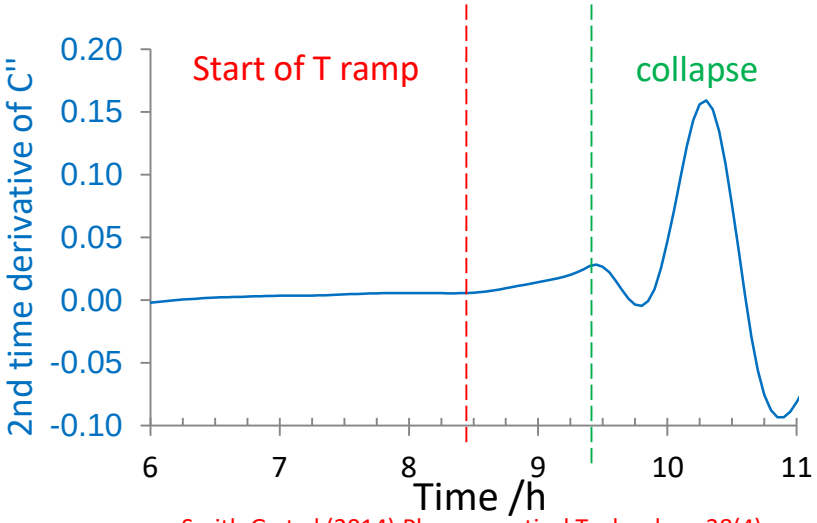
Product characterization:
Glass transition, Eutectic Crystallization, Collapse

Smith G et al (2013) Eur J Pharm Biopharm 85 (3)1130-40



Arshad MS et al (2014) Eur J Pharm Biopharm 87(3):598-605





Smith G et al (2014) Pharmaceutical Technology 38(4)

A suddenly change in the capacitance demonstrates the collapse of cake