SANOFI

Measurement of Residual Moisture Content by Non-Invasive Methods

Timothy R McCoy - **Global Lyophilization CoE, Sanofi R&D**

Objective

- **PAT in Freeze Drying Overview**
- **Residual Moisture in Drug Product post FD Process and on Stability**
- **Sources & Measurement of Residual Moisture**
- **Scale up Implications – Process Impact & Testing Burden**
- **Case Study – comparing Near IR to Frequency Modulated Spectroscopy**

Process Analytical Technology (PAT)

- **Definition (US FDA)*:**

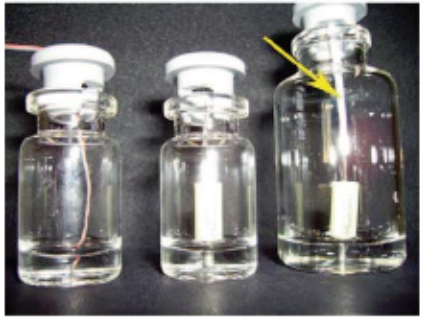
Mechanism to design, analyze, and control pharmaceutical manufacturing processes through the measurement of Critical Process Parameters (CPP) which affect Critical Quality Attributes (CQA).

***Guidance Document**
PAT—A Framework for Innovative Pharmaceutical Development, Manufacturing, and Quality Assurance (FDA, 2004)

- **Relative to Freeze drying – Process Monitoring or true PAT?**
  - Operations sites want defined cycle times for supply chain planning

FDA Guidance clear that PAT must be used in manufacturing of pharmaceutical products

Lyophilization Process Development



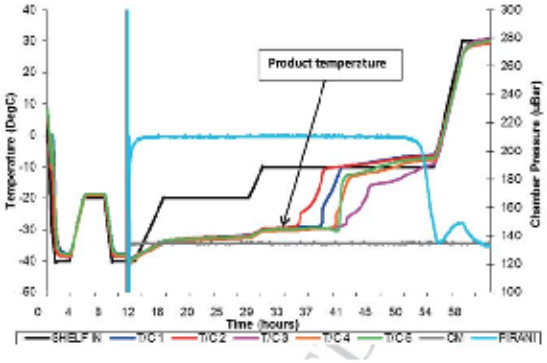
Formulation, Dry layer Resistance (Rp); Physical & Biophysical Characteristics

Vial Heat Transfer Coefficient (Kv) & Process Performance

LYO Model & Small Scale Development

Process Modelling & Monitoring Tools at both Pilot & Large Scale

Commercial Lyophilization Process



Lyophilization Cycle

Monitoring of Key Variables/ Attributes

Variable or Attribute	Notes
Product Temperature	Most important parameter in Freeze Drying Process
Mass Transfer – Sublimation, Desorption	Primary & Secondary Drying performance, endpoint, Rp, Kv, load factor, Chamber vacuum Control
Physical Characteristics	Monitoring Phase Transitions in Real Time
Residual Moisture	Target as many samples tested as possible in scale up to understand variability, then reduce for PPQ/ Commercial

Process Analytical Technology in Freeze Drying

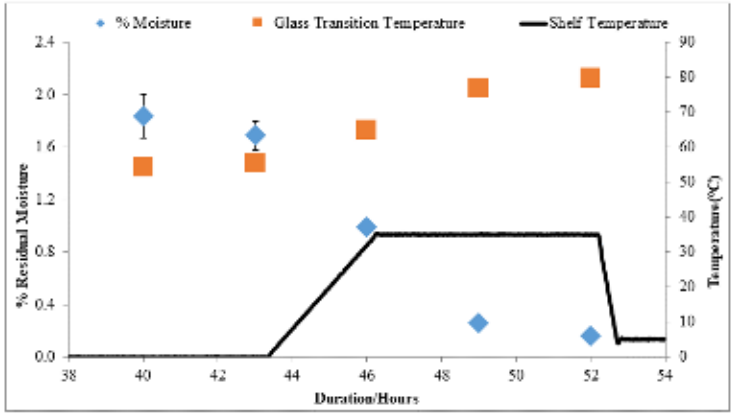
Application	Technology	Notes
Product Temperature Measurement	Thermocouples (wired, wireless) Temperature Sensors (e.g. Tempris, LyoDEA) RTDs (e.g. ELLAB)	Destructive/ Non-Destructive
Primary & Secondary Drying	Pirani Gauge v Capacitance Condenser Temperature Shelf Inlet v Outlet MTM TDLAS Mass Spec IR Monitoring (Camera) Raman	Sublimation rates, Cake Resistance, endpoint point; Desorption rates, Secondary Drying profile Represents entire batch, not location specific, does not differential between location; Silicone Oil but also vapor (quite sensitive for secondary drying) Monitoring of drying rates, Kv – limited to vials near sensor
Residual Moisture Analysis	Near-Infra Red (NIR) Frequency Modulated Spectroscopy (FMS)	Non Destructive – a way of measuring residual moisture either on bench or inline
Phase Transitions	Heat Flux Sensors LyoDEA	Possible to monitor phase transitions during FD process

Application	Technology	Notes
Ice Nucleation Control	Ice Fog, Depressurization, FreezeBooster®	Control of crystal size, orientation and degree of supercooling

Residual Moisture of Lyo Drug Product

Why is it important?

- **Directly related to the physical state of the Lyo DP**
  - Homogeneity v Heterogeneity – vial to vial: consider location and ice nucleation; within a single cake: phase separation, drying process
- **Residual Moisture v Tg – inverse relationship**
  - Opportunity for excipients to crystallize once Tg is lower than storage temperature



%RM & Tg as a function of Secondary Drying

Sources of Residual Moisture

Lyo Process and Elastomer Contribution on Stability

Sources	Key Information	Notes
Lyophilization Process	All steps - Freezing, Primary & Secondary Drying	Residual Moisture at T0 – contribution due to Lyo process only, not from stopper
Stopper contribution	Transmission through & transfer from stopper	Residual Moisture on Stability - Increase in moisture from source, change in physical state (drop in Tg) - Opportunity for further phase transitions if Tg< Storage temp e.g. 40°C

Measurement of Residual Moisture of LYO DP

Method	Sample Management	Notes
TGA	Loss on Drying, open exposure of sample	Manipulation of Sample; Lyo cake hydroscopic, impact results; Measurement includes volatiles, everything that evaporates
KF (Coulometric, Volumetric)	Direct addition of sample to organic solvent or dissolve in organic solvent & then add	Manipulate sample, sample prep, time
KF Oven	Place complete vial in oven and apply heat; specific to moisture only (nitrogen carrier) Completely closed, sample not manipulated	Sample prep, time, method (apply head – design very carefully considering Tm)
NIR, FMS	Non invasive No Sample manipulation (also O2 Headspace)	Calibrated using KF, can be assessing global moisture Limitations include cake appearance impact, phase changes impact on headspace, method variability v KF

Challenges in Tech Transfer & Scale Up

Increased Sample Numbers for Residual Moisture Analysis

Scale	Batch Size*	Samples For Tech batches**	# shelves***
Bench	50 vials	3-5 vial	1
Pilot	100-300 vials	10 vials	3
Clinical GMP	1000-5000 vial	20-40 vials	5
Commercial GMP	30000 vials +	60+ vials	10

* Lyo Capacity Dependent
** Dependent on company practices and interpretation of regulatory guidance
*** Can vary depending on Lyo design, company protocol/ policy; provided as an example

As we scale up, ideally we should have a robust picture of residual moisture heterogeneity inter and intra batch; important to demonstrate comparability/ scalability

Lyophilization Scale Up

Considerations on Impact to Residual Moisture

- **Environment – lab (unclassified) v class 100**
  - Particle Levels
  - Degree of supercooling and Rp can be impacted
- **Heat Transfer Coefficient, Kv – need to adjust cycle to target product profile**
- **Batch size impact – differences from lab and at scale between min to max loads**

Fair to make an assumption that the Lyophilization Cycle has been developed appropriately

Karl Fischer vs Non Destructive Analysis

High Level Comparison in Moisture Determination

	Karl Fischer	NIR, FMS
Sample Handling	Destructive	Non Destructive
Sample Recycling	Sample disposed post testing	Sample can be used for another test
In line/ Real Time Analysis	No in line option	In Line possible
Sample Number	Small Sample Numbers	Larger Sample Numbers up to 100% batch
Operational	Time Consuming	Fast and Efficient

- Method Variability better for KF, but can improve NIR, FMS with larger sample set (as low as 0.1%)
- Objectives is not to replace KF as release method, but use non destructive techniques for technical/ scale up batches to increase data set

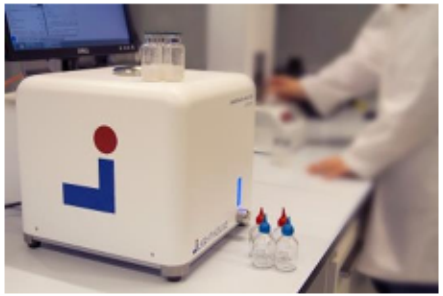
Non-invasive Moisture Determination

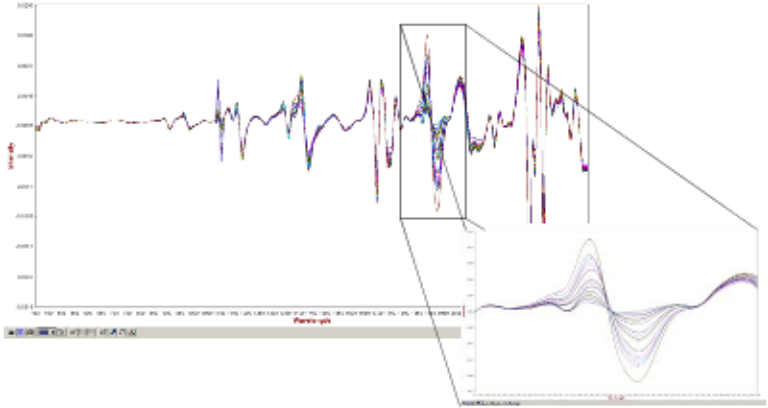
Benefits Include

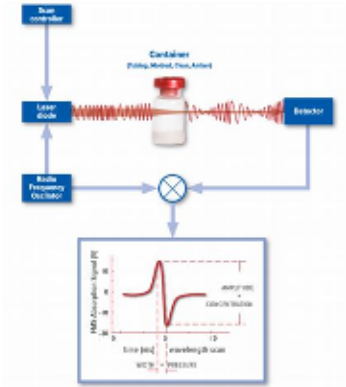
- ✓**Lyophilization process characterization by moisture mapping of freeze-dried products**
- ✓**Improved compliance with capability of up to 100% in-process checks**
- ✓**Significantly reduced number of drug product units for release testing based on comprehensive assessment during scale up**
- ✓**Real time analysis of Residual Moisture (in line)**
- ✓**Deterministic CCI methodology**

Residual Moisture Technology

Near IR (NIR), Frequency Modulated Spectroscopy (FMS)







Headspace Method
Modulation techniques result in 10,000x increase in sensitivity compared to first order absorption techniques such as NIR

Experimental

Material & Methods

Description	Details
Protein	Model Protein
Formulation	Sucrose based
Primary Packaging	Schott 20cc Type 1 glass tubing vial Stopper – West Novapure S87-I 4432/50 Lyotec, B02 Fill volume (Vfill) A = 5.3 mL
Primary Packaging Prep	Vial washed and dried Stopper prep – washed with WFI, dry at 100°C (table later in slides)
Sample Preparation	Samples – Lyo product & stopper Generated at different residual moisture levels in cake and stopper, exposed to 40°C
Residual Moisture Measurement	Karl Fischer (control), FMS & NIR

NIR System Details

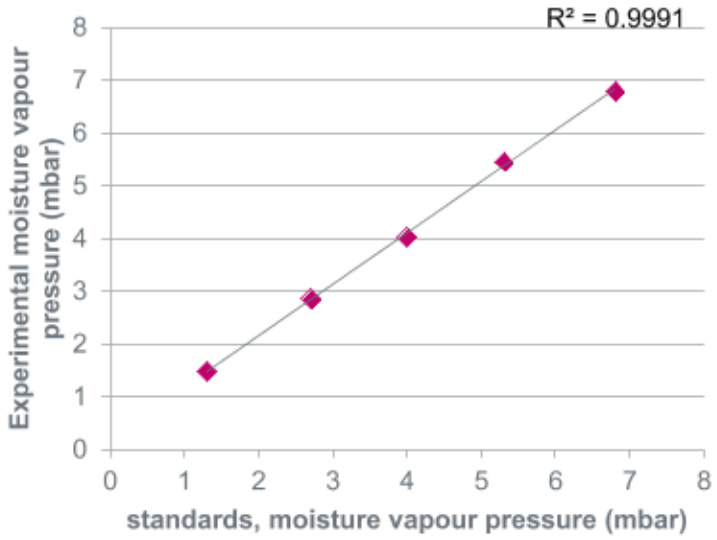
Description	Details
System	Metrohm model XM-1000 with XDS rapid content analyzer
Measurement	Placing the vial on the glass platform and scanning from the bottom of the vial into the lyo cake
Detection	Linear regression model at 1887 to 1960 nm (based on Karl Fischer moisture values)
Calibration	Ceramic discs, with different reflectance, placed on the glass and scanned Internal ceramic reference standard that is scanned at start up

FMS System Details

Description	Details
System	Lighthouse model FMS 1400
Measurement	Laser (1400 nm) passed through the headspace of a sealed drug product vial
Detection	Laser at 1400nm (laser absorption spectroscopy)
Calibration	Standard calibration vials (hermetically sealed vials) are supplied by vendor calibration check for 5 standard vials (hermetically sealed) in the range of 1mbar to 7mbar of moisture vapor pressure

FMS Calibration

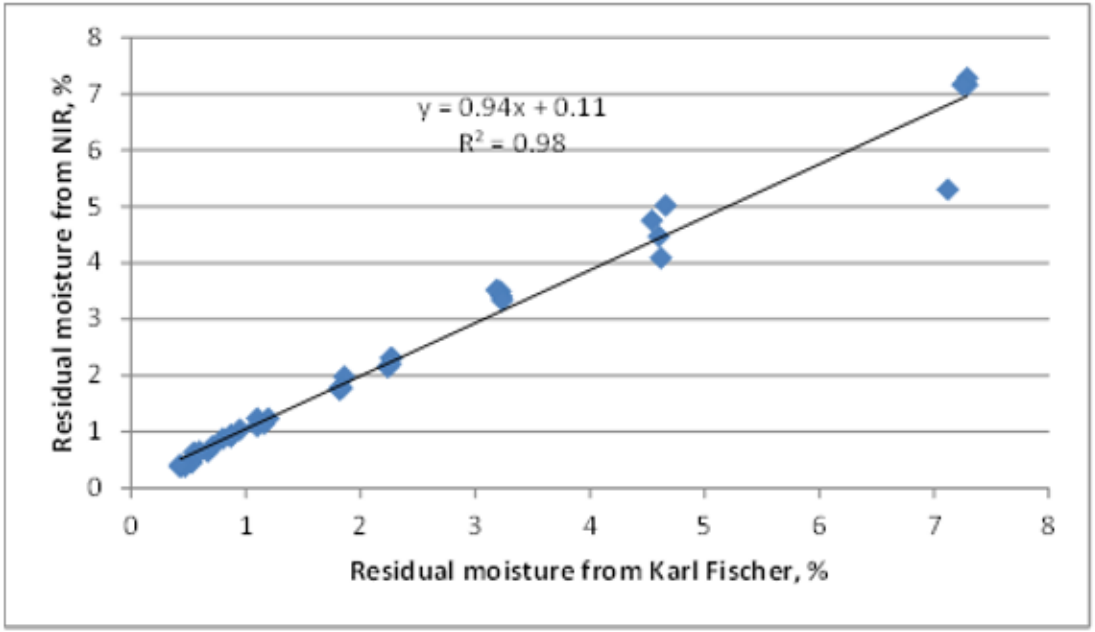
- **R2 >0.99 linear correlation is obtained.**
- **A full calibration is complete prior to any measurement.**

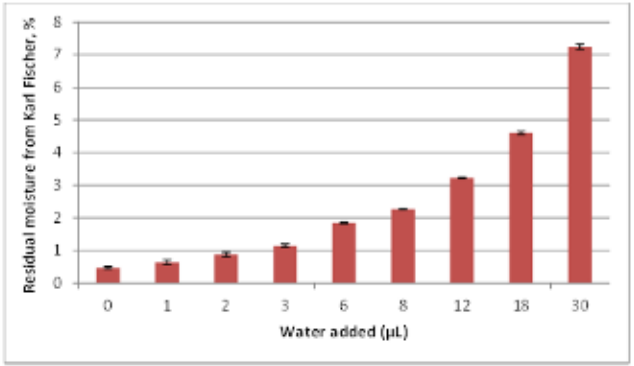


Typical calibration check

NIR Residual Moisture Calibration

Calibration Set & Correlation of NIR vs KF (Water Addition)





Generation of moisture samples by water addition

Water addition amounts for NIR vials based on lyophilized cake weight to achieve theoretical desired increase in residual moisture.

Stopper Moisture Study

- **Assess any potential moisture impact to Drug Product from the Stoppers**
- **Purpose of such Studies:**
  - Define the acceptable range of moisture
  - Prepare stopper subsets with varying moisture and assess moisture over a period of time
- **Formulation lyophilized as per specific protocol using stoppers with various moisture content.**
- **Samples placed on stability (real time and accelerated stability condition, 40°C) and checked at different time intervals.**

Force Degradation Study

Experimental Design

Conditions	Stability Timepoint (T = months)				
	0	3	6	9	12
2-8°C	X	X	X	X	X
25°C 60% RH		X	X	X	X

X= product sample taken (n=3)

- **Drug Product exposed 80°C to drive moisture our of stopper into product**
- **Stoppers prepared at different % RM levels (next slide)**
- **All timepoints contained all stopper subsets and n=3 replicates**

Stopper Preparation

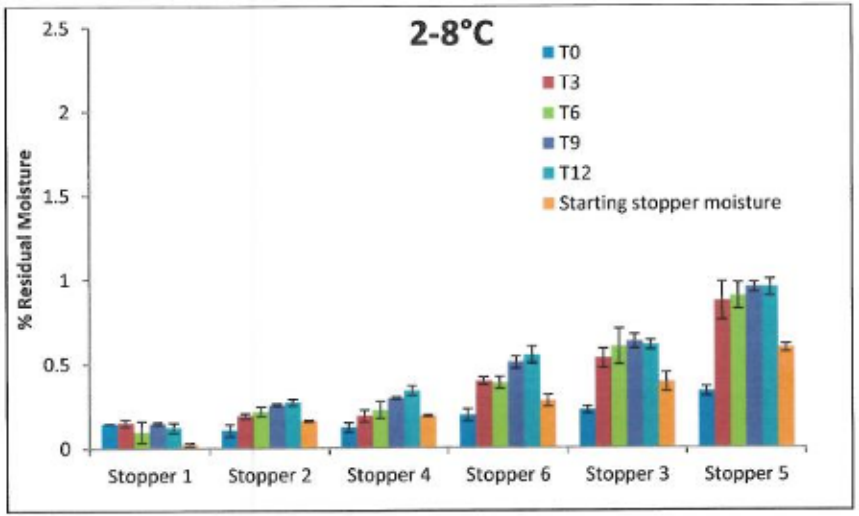
Force Degradation Study

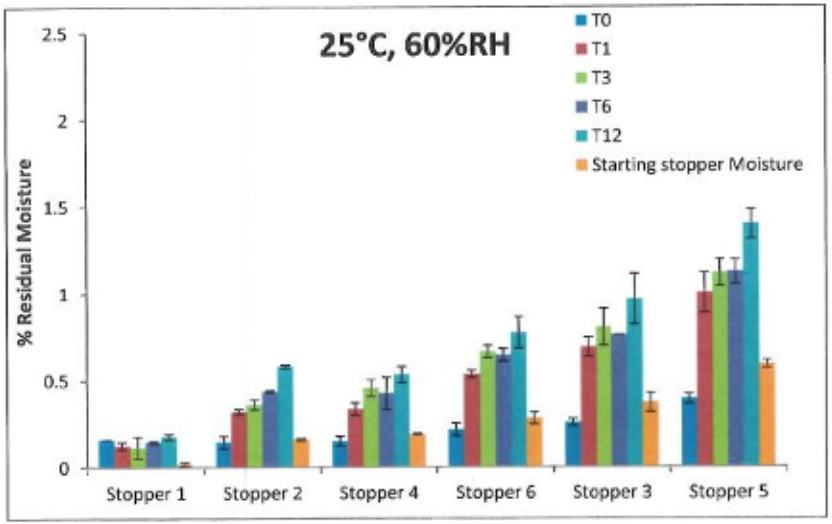
Stopper Subset	Method of Stopper Preparation	% Residual Moisture (Ave)	SD (n=3)
Subset 1	72 hours drying at 100°C	0.02	0.01
Subset 2	2.5 hours at 50°C followed by 1.5 hours air drying	0.16	0.01
Subset 3	Immersed in water, 2.5 hours at 120°C followed by drying with tissues	0.39	0.06
Subset 4	No additional treatment (untreated stopper)	0.19	0.01
Subset 5	Immersed in water, 4 hours at 120°C followed by drying with tissues	0.59	0.02

Force Degradation Study

Residual Moisture Measurement of Product by FMS

- **Conditions 5 & 25°C**

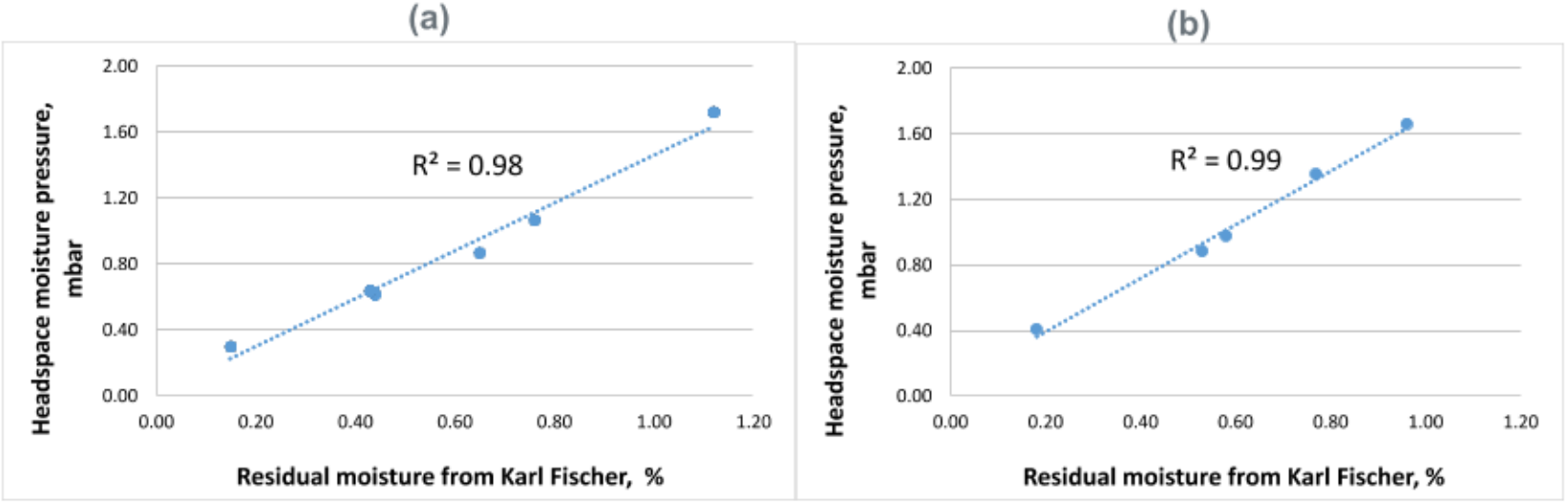




Product RM Data function of Stopper Moisture & Stability Timepoints at 2-8 & 25°C measured by FMS

FMS vs KF Correlation – Force Degradation

Drug Product %RM Correlation at 25°C after 6 and 12 months



Correlation of FMS headspace moisture pressure and percent residual moisture for stopper moisture samples **(a)** 25°C, 60% RH 6 months, **(b)** 25°C, 60% RH 12 months.

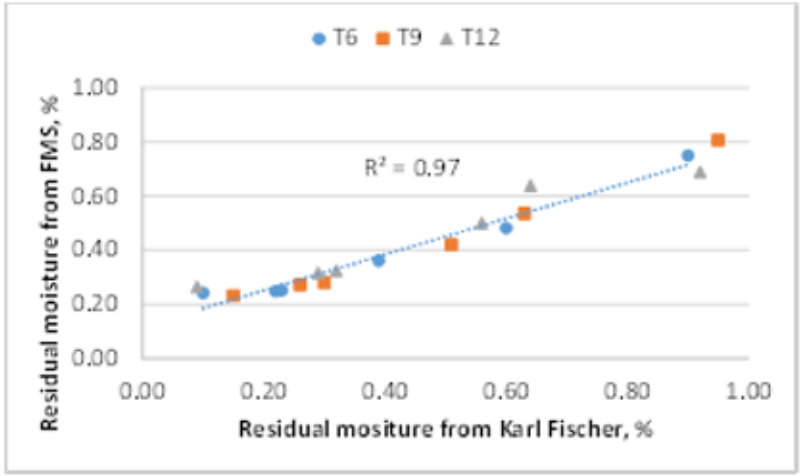
$$RM = (mp + 0.08)/1.82$$

Where RM – residual moisture and
mp – FMS moisture pressure

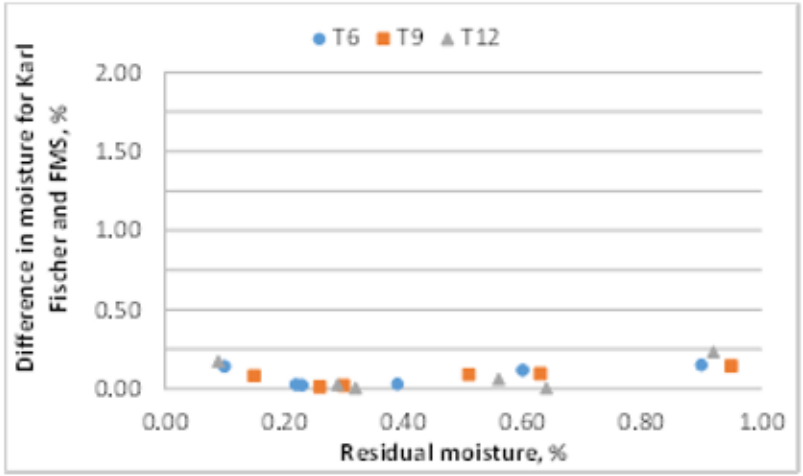
Residual Moisture Calculation - FMS

FMS v Karl Fischer

(a)



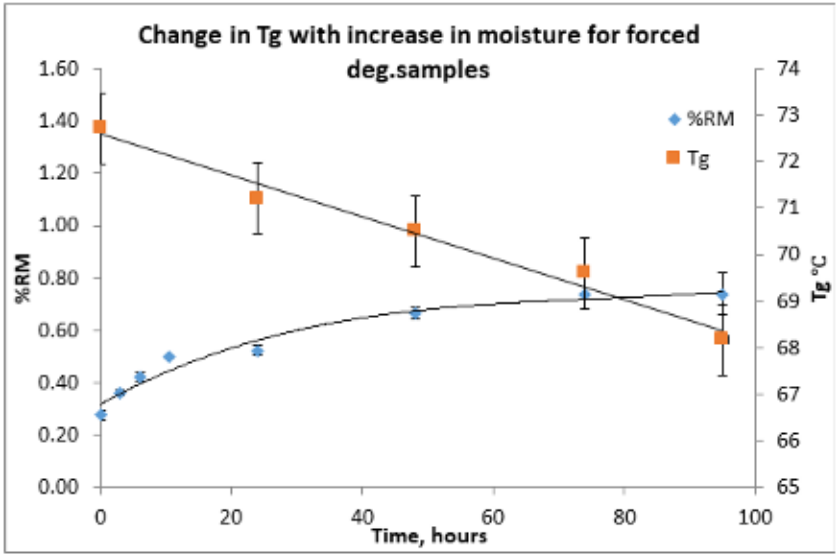
(b)



The experimental Karl Fischer residual moisture and calculated FMS residual moisture values for all 2-8°C. Calculated and experimental percent residual moisture values **(a)** and the difference values of this regression **(b)**.

Residual Moisture v Tg of Lyo Drug Product

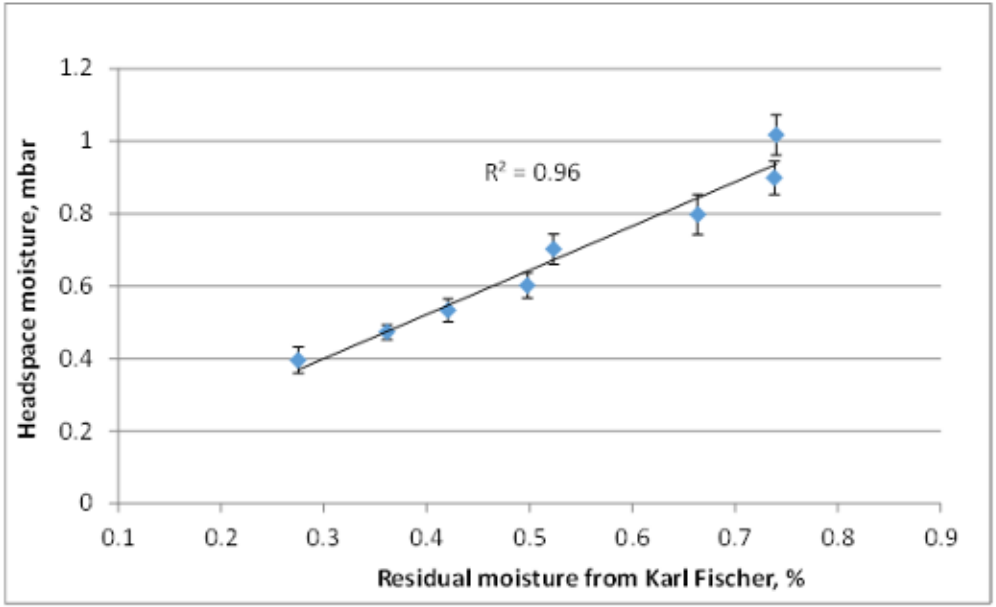
%RM Measured by FMS



Force Deg Study – exposing Product to 80°C to study moisture transfer from stopper over short period

Correlation Residual Moisture

FMS v KF method – Product Samples by Force Degradation



Force Deg = Product held at 80°C for 4 days to drive moisture out of stopper

Use of Karl Fischer v Non Destructive

Conclusions

- **Karl Fisher Gold standard for determining % RM in Lyo cakes**
- **The overall accuracy of KF is better (not shown here), but benefits of headspace technology involve:**
  - Only few vials (5-10) vials out of 30-60K vials will be checked for moisture - not representative
  - Safety - related to chemicals used in Karl Fisher - chemicals are CMR chemicals, there is a safety risk in mix-up of Hydranal with other acids which is typically used for cleaning Karl Fisher instrument.
  - KF can be time consuming
- **Often the moisture levels are so below than specs, 0.1-0.2%RM inaccuracy is not important**

Use of Non Destructive Analysis

Conclusion

- **Possible to determine %RM for hundreds of vials (in line or post inspection) – maximizing understanding of variability**
- **High throughput (scanning time in seconds)**
- **Non destructive**
- **Accuracy to be improved with larger data set, can be improved in the region of 0.1%**
- **Cake shrinkage/collapse will impact NIR**
- **Phase Changes in DP can impact headspace (FMS analysis)**

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Thank You!

On behalf of Richard, Kumar and myself!!

Questions?

Timothy.mccoy@Sanofi.com