The Mechanisms by which TBA Enhances the Freeze Drying of Pharmaceuticals

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Use of Organic Solvents in Freeze-drying

- Objectives:
  - Freeze drying of water insoluble products
  - Freeze drying of extreme water-unstable products
  - Increased rate of sublimation and shorter drying cycles
  - Improved dry product stability
  - Improved reconstitution time
- Solvents used: (Generally with water)
  - Tertiary Butyl Alcohol: e.g., few products
  - Dimethylsulfoxide (DMSO): e.g., intravesicle Rimso-50
  - Isopropyl alcohol
  - Ethanol, e.g., Chemo agent

Physical Properties of commonly used organic solvents used in lyophilization

Property	Water	T-BA	**DMSO**
Mol. Wt.	18.02	74.12	78.14
Melting Point (C°)	0	25	19
Boiling Point (C°)	100	82	189
Density (g/mL) 25 °C	0.997	0.7812	1.096
Vapor Pressure (mmHg)	23.78	41.25	0.60
Viscosity (mPa.S) 20 °C	1.00	3.62	2.20
Heat of fusion (cal/gm)	80	22	41
Heat of vaporization (cal/gm)*	580	130	162

Ideally, the solvent should have:

- ***high volatility,***
- ***freezable (form an eutectic),***
- ***condensable on the condenser,***
- ***non-toxic (even if removed at the end of the process),***
- ***non-reactive or inert, and***
- ***aid the drying process***

*** Temp dependant**

Toxicology/safety

- Similar as other Class 3 solvent : 50 mg/day
- OSHA: Daily exposure limit 300 mg/M^3
- LD_{50} (mg/Kg) –
    - IV mouse: 1538
    - IP Mouse: 399
    - Oral: Rat 2743, Rabbit 3559
    - Skin: rabbit >2000, G. Pig > 2000
- Reference: “Tertiary-Butanol: a toxicological review”, by McGregor D., Crit Rev Toxicol. 2010, 40(8), 697-727.
- Very low levels in lyophilized product: e,g, 0.2 mg/Vial

Some Early Experiments Using tBA

M. Kamat, Ph.D. Thesis, UofKy 1989

EFFECT OF SOLVENTS

Effect of solvents (5%, w/v) on freeze drying sugar solutions.

System	Shelf Temp (°C)	Remarks
Sucrose alone	0	Collapse
Sucrose + methanol, ethanol, IPA, acetone nBA, dioxane	0	Boiling of solvent Collapse
Sucrose + tBA	30, 45	Good
Lactose alone	0	Collapse
Lactose + methanol ethanol, IPA, acetone nBA, dioxane	0	Boiling of solvent Collapse
Lactose + tBA	30, 45	Good

Except in the case of tertiary butyl alcohol (tBA), all other solutions failed to freeze-dry. Severe bubbling of the co-solvents and collapse occurred during the early drying phase. Solutions containing tBA, however, resulted in complete drying and yielded good cakes. All the co-solvents, except tBA, have freezing points well below the temperatures encountered during the freeze drying process. Visual observation of solutions containing tBA during the freezing stage indicated that tBA froze in characteristic needle-shaped crystals much before ice was formed. Phase diagrams of tBA-water and tBA-water-sucrose also indicated that tBA was frozen at the drying temperature during the cycle. Presence of such solidified tBA in the frozen solutions and its sublimination without passing through the liquid phase may have strengthened the matrix and thereby prevented collapse.

M. Kamat, Ph.D. Thesis, UofKy 1989

FREEZE DRYING BEHAVIOR OF SUGAR SOLUTIONS AT VARIOUS TEMPERATURE

	Freezing Pattern	Collapse Temp. oC	Max. Shelf Temp. oC	Drying Rate, g/Hr
Sucrose alone	Spont.	-28	-30	0.21
Sucrose + 5% tBA	Slow needles	-21	+30	0.60
Sucrose + 10% tBA	Slow needles	-20	+30	0.67
Lactose alone	Spont.	-23	-15	0.19
Lactose + 5% tBA	Slow needles	-22	+30	0.70
Lactose + 10% tBA	Slow needle	-21	+30	0.79

In the presence of tBA, the initial drying rate was 3 times faster at 30^{o}C, however only the solutions which contained tBA survived at this termperature. With lactose the corresponding increase in drying was almost 3.5 times faster.

The freeze drying cycles of several proteins have been accelerated with tBA. These include hemoglobin and other antibodies.

M. Kamat, Ph.D. Thesis, UofKy 1989

PHYSICAL CHARACTERISTICS OF FREEZE-DRIED HEMOGLOBIN

Formulation	Shelf	Cake color	Reconstitution
Hb alone	0°C	Dark brown	Difficult
Hb + NaCl (0.9%)	0°C	Dark brown	Difficult
Hb + tBA (5%)	0°C	Dark brown	Difficult
Hb + Sucrose (5%)	0°C	Bright red	Easy
Hb + Sucrose (5%) + tBA (5%)	0°C	Bright pink	Easy
Hb + Sucrose (5%) + tBA (5%)	30°C	Bright pink	Easy

M. Kamat, Ph.D. Thesis, UofKy 1989

CHEMICAL PROPERTIES OF FREEZE-DRIED HEMOGLOBIN

Formulation [a]	Hb content (%, w/w)	MetHb content (%, w/w)	Moisture (%, w/w)
Hb alone	52	46	5.3
Hb + NaCl (0.9%)	39	60	5.2
Hb + tBA (5%)	65	32	3.1
Hb + Sucrose (5%)	97	< 1	1.6
Hb + Sucrose (5%) + tBA (5%)	97	< 1	5.1
Hb + Sucrose (5%) + tBA (5%) [b]	98	< 1	3.0

a: Drying Temperature = 0°C except b = +30°C

M. Kamat, Ph.D. Thesis, UofKy 1989

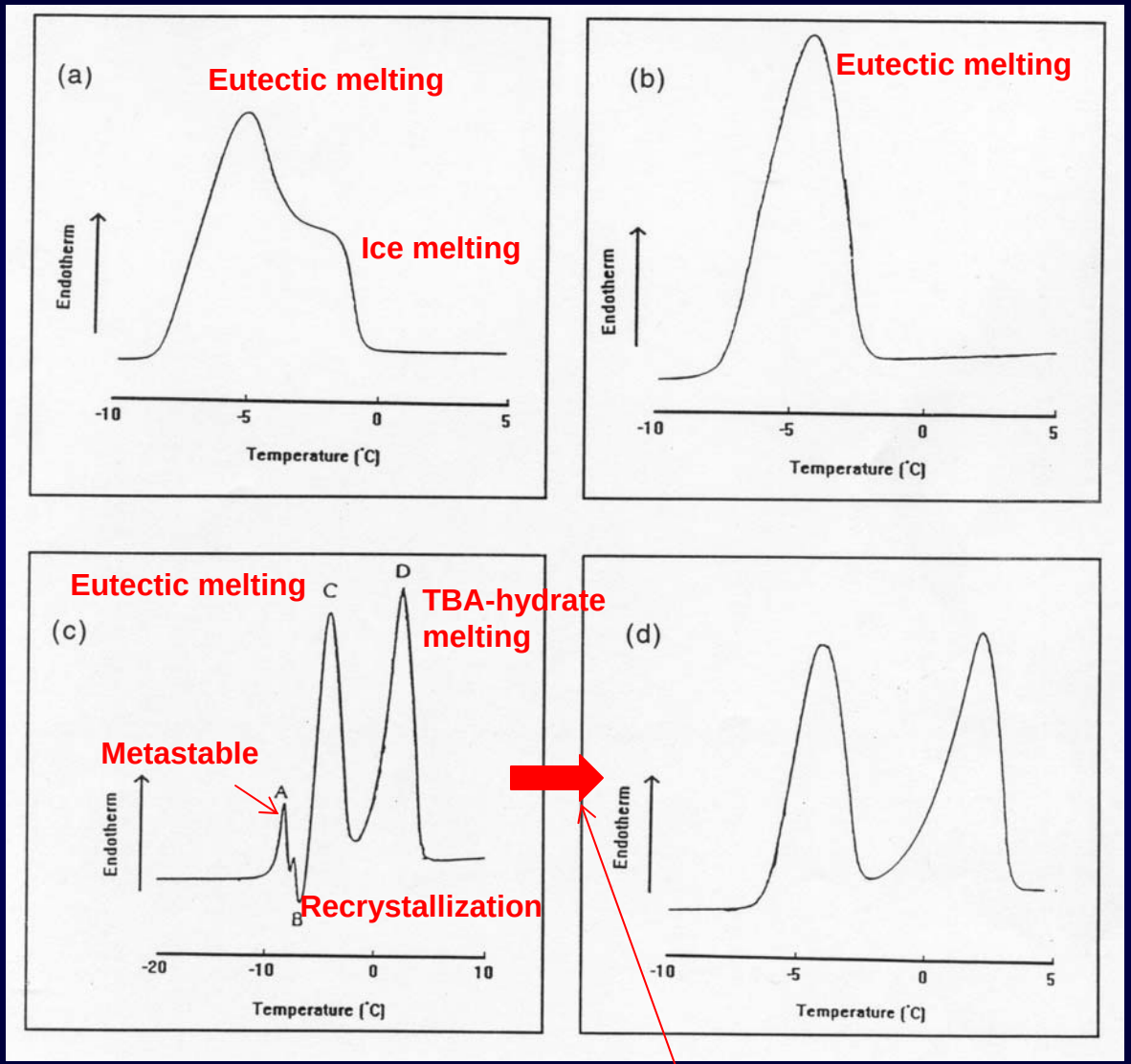
tbA-water freezing behavior

Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 484-490, 1995

DSC Warming Thermograms for tBA-Water Mixtures

(a): 15% TBA

(b):20% TBA



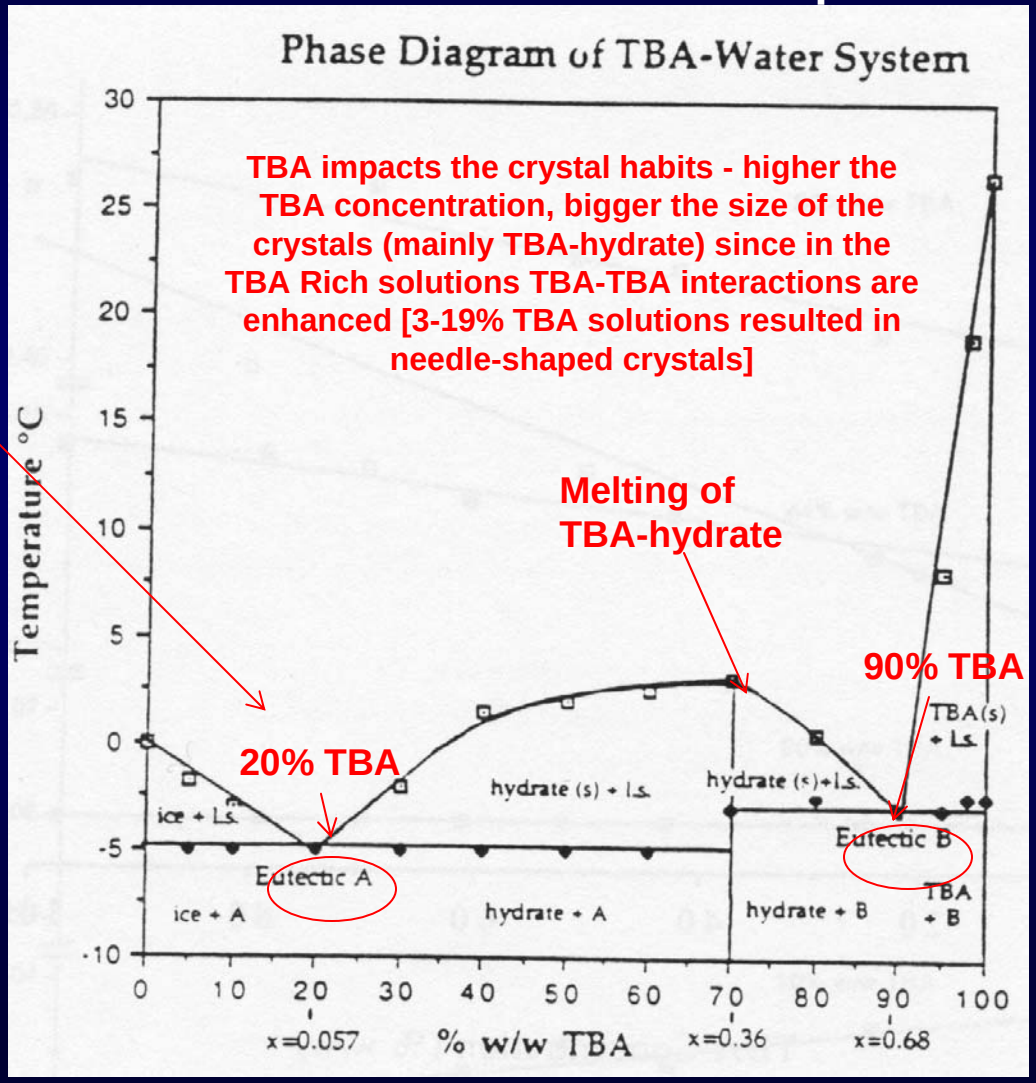
(c):
Endotherm A: melting of metastable eutectic
Exotherm B: recrystallization of metastable eutectic to form stable form
Endotherm C: melting of eutectic
Endotherm D: melting of TBA hydrate

(d) : Thermal treatment of 50% solution at -7°C to eliminate metastable states

Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 484-490, 1995

Phase Diagram of TBA-Water System

Phase diagram of the TBA-water system plotted on % (w/w) basis. Mole fractions are listed for Eutectic A and B and the pure TBA-hydrate.

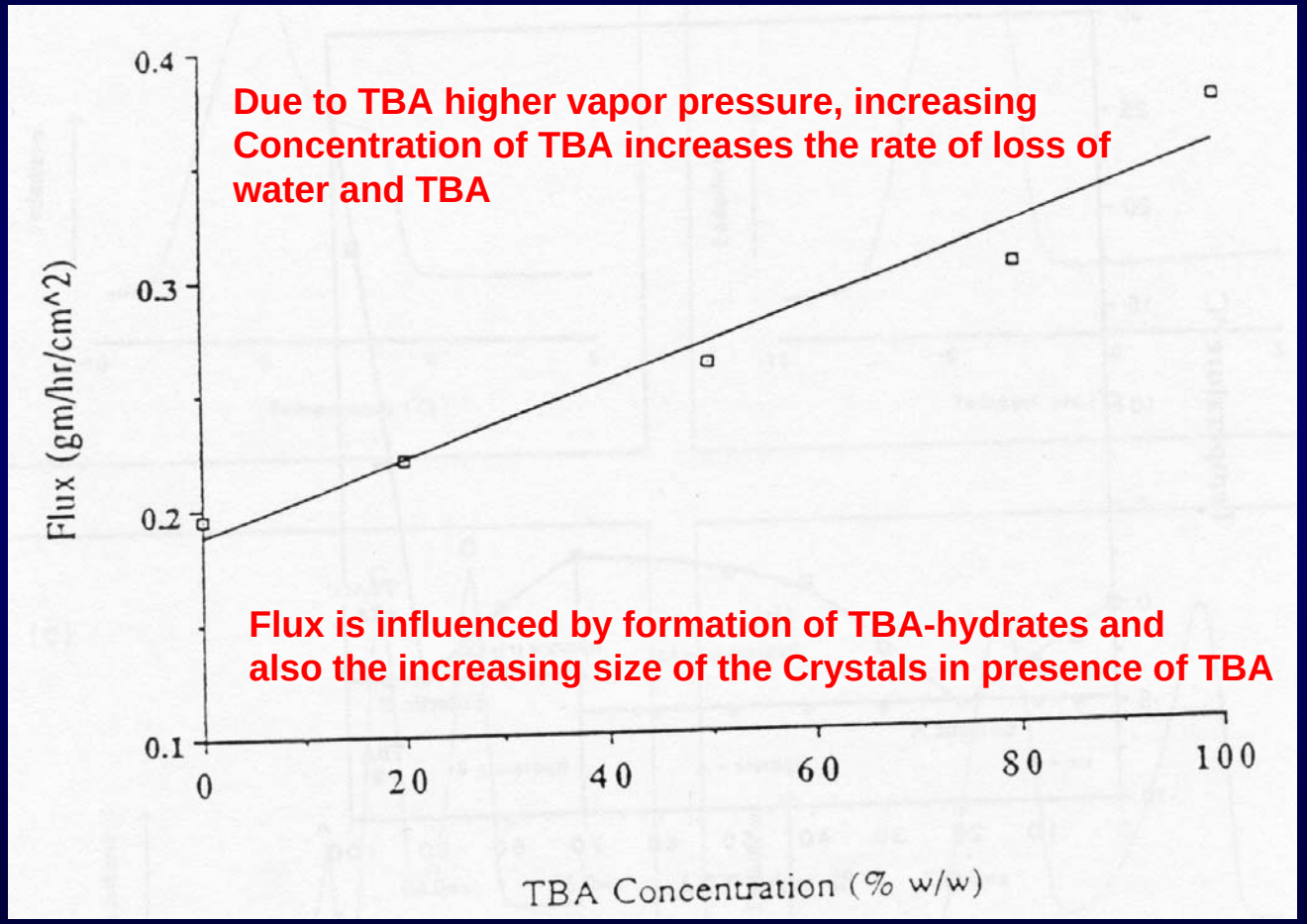


Eutectic A at 20% TBA - Both water and TBA Crystallizes out upon cooling

Eutectic B at 90% TBA - Both TBA and TBA-hydrate Crystallizes out upon cooling

Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 484-490, 1995

Flux of TBA/Water Mixtures as Function of TBA Concentration (Shelf temp 0°C and Pressure 100 mTorr)



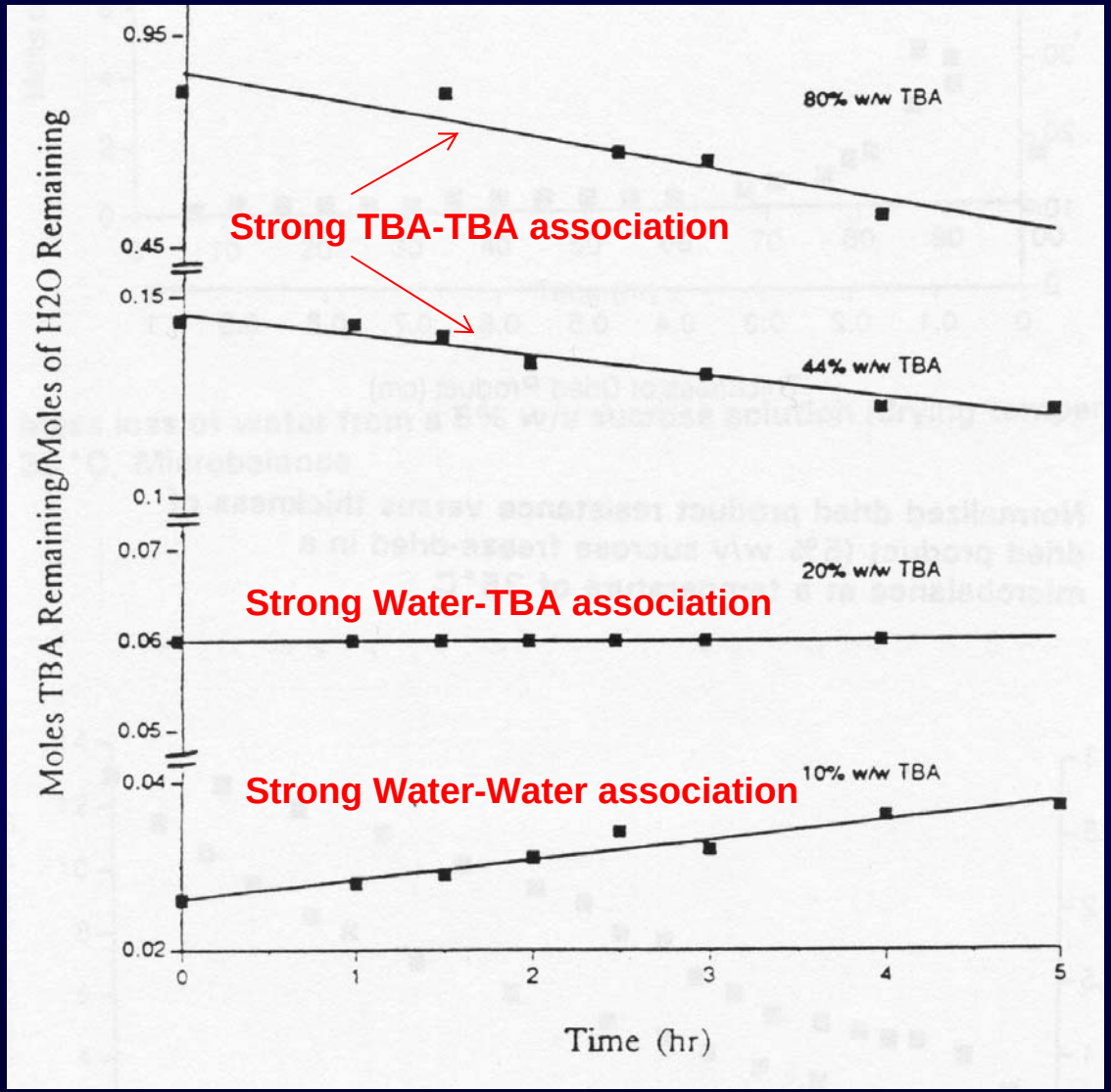
Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 484-490, 1995

Ratio of TBA to Water as Function of Time for 10%, 20%, 44%, and 80% TBA Solutions

TBA molecules sublime faster at 44% and 80% since they fall in the TBA rich region of the phase diagram

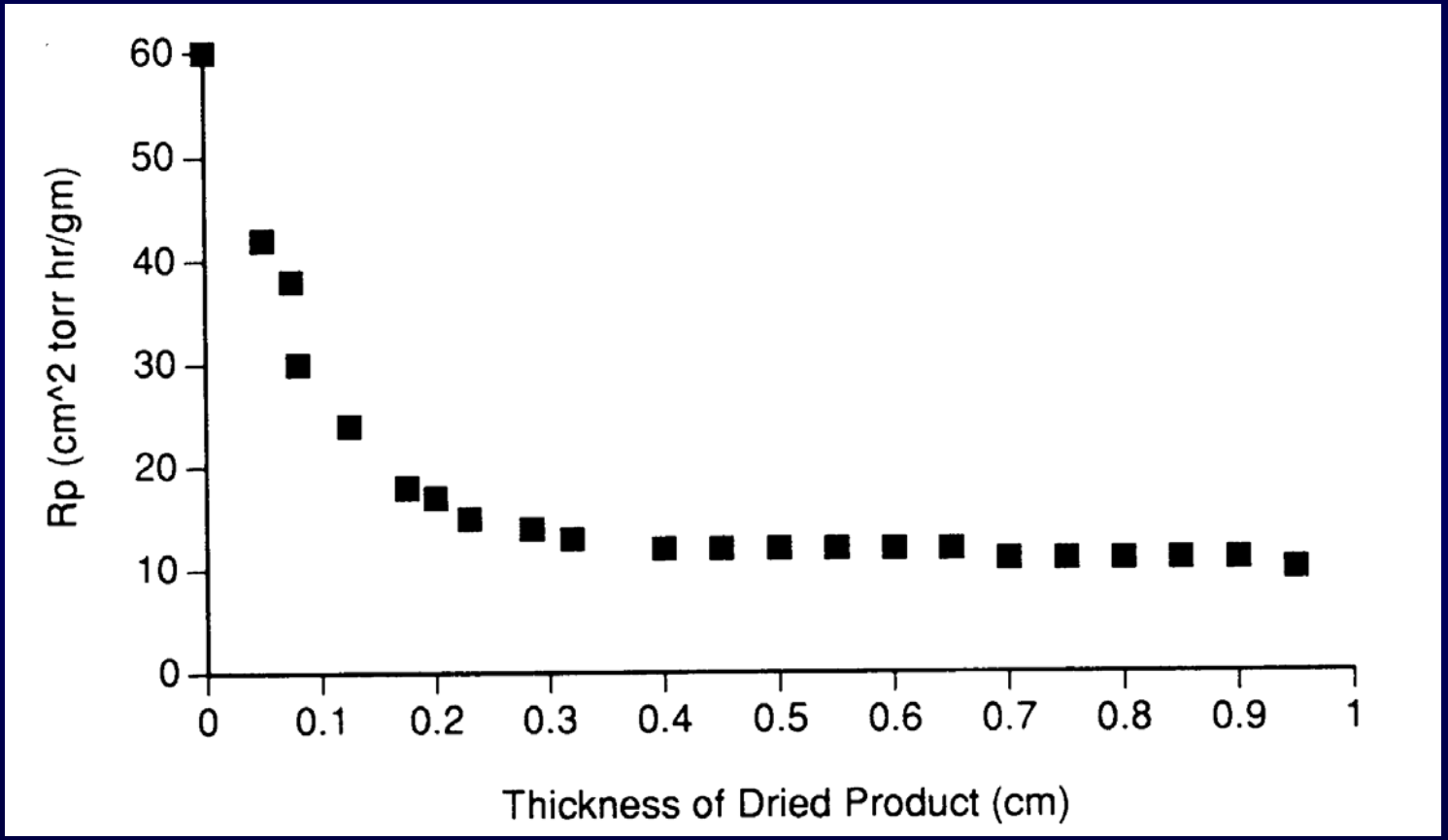
Water and TBA molecules sublime at the same rate at ~20% TBA concentration

Water molecules sublime faster at ~10% TBA concentration, since they fall in water rich region of the phase diagram



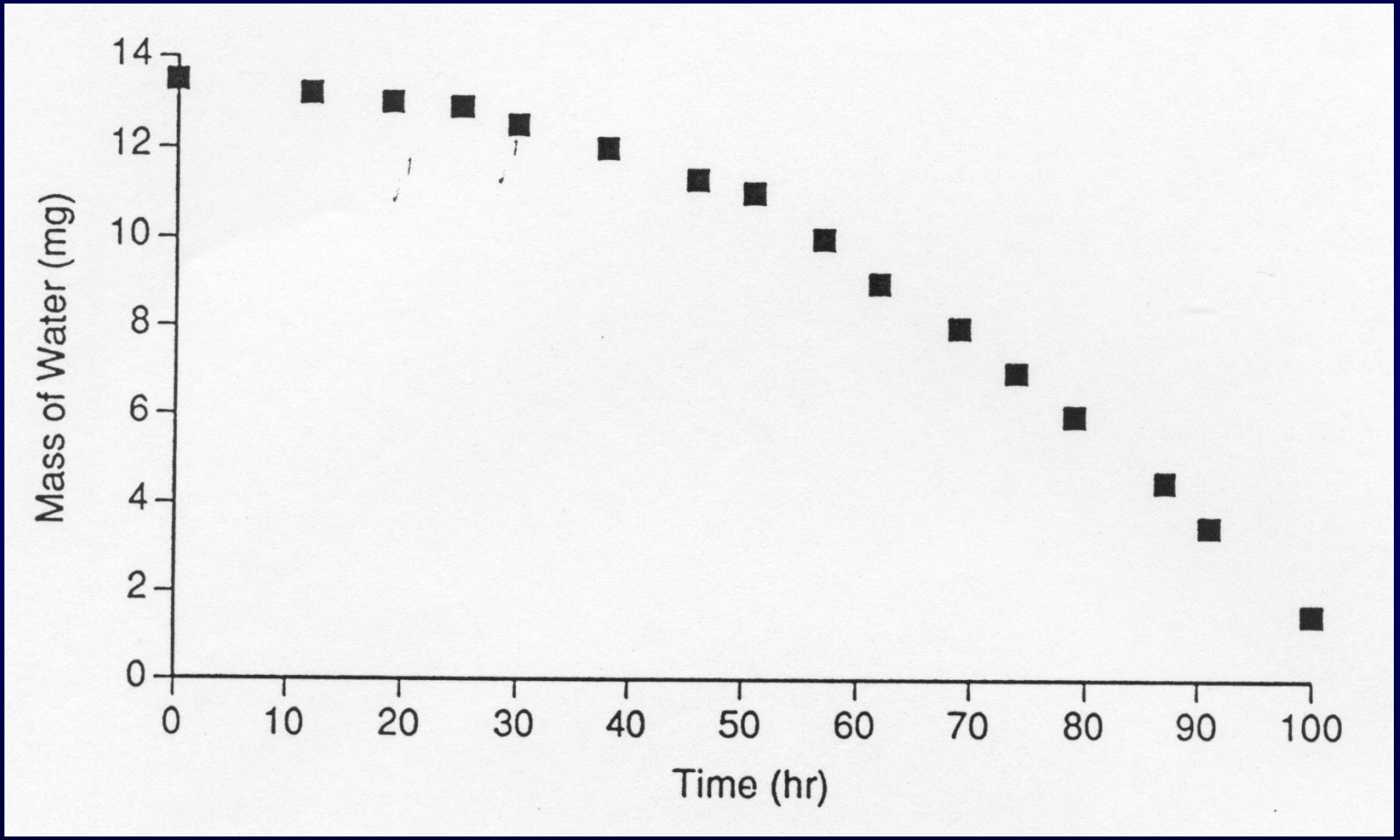
Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 484-490, 1995

Normalized product resistance versus the thickness of dried product for 5% w/v sucrose solution (drying temperature -35°C. Microbalance)



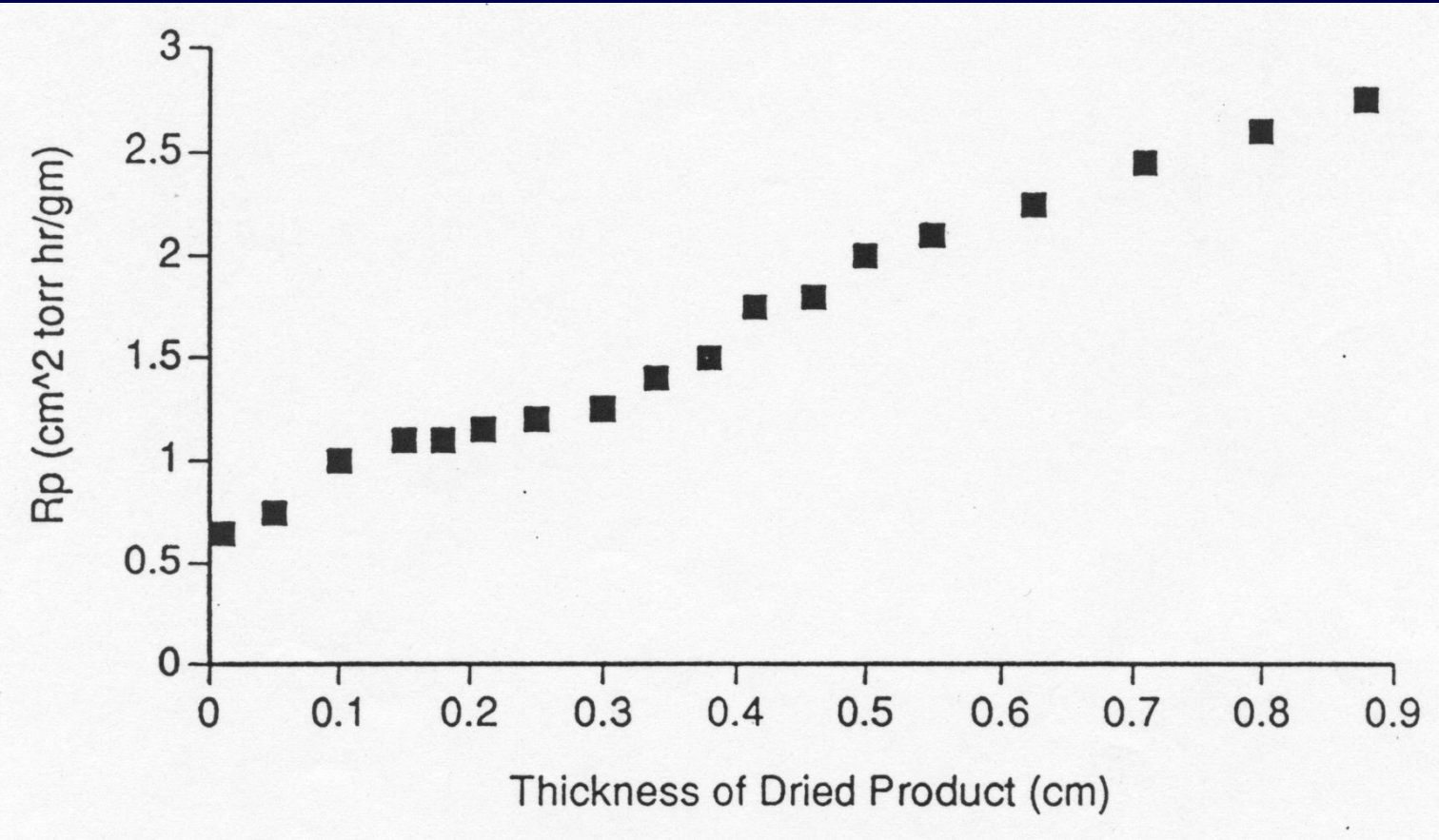
Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 491-495, 1995

Mass loss of water from a 5% w/v sucrose solution (drying temperature -35°C. Microbalance)



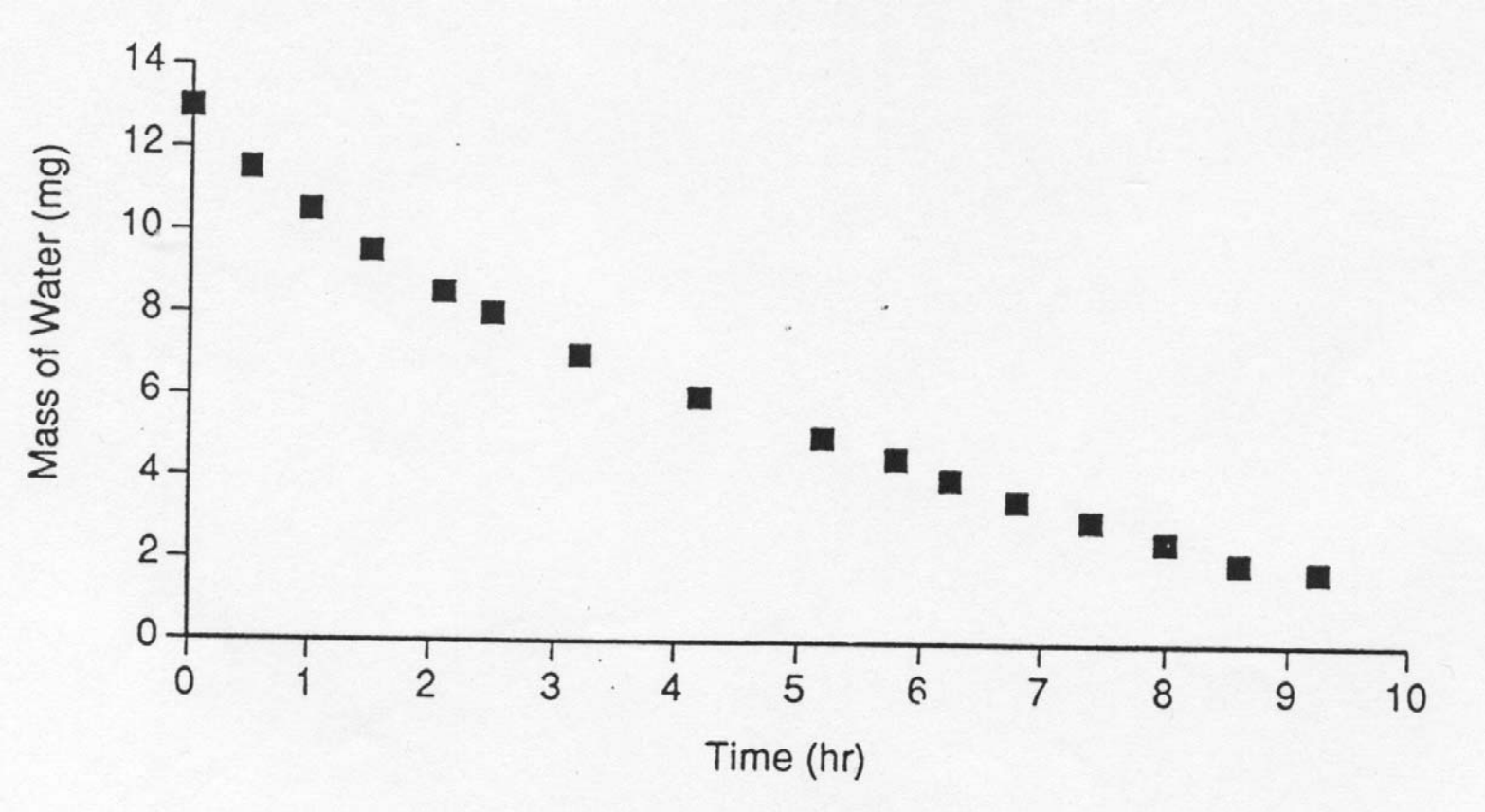
Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 491-495, 1995

Normalized dried product resistance versus thickness of dried product (5% w/v sucrose containing 5% w/v TBA freeze-dried product in a microbalance at a temperature of -35°C)



Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 491-495, 1995

Mass loss of water from a 5% w/v sucrose solution containing 5% w/v TBA (drying temperature -35°C, Microbalance)



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Summary: Freeze-drying of tBA/Sucrose/water

Reference: K. Kasraian and P. Deluca; Pharm Res., Vol 12: 4, 491-495, 1995

- Addition of 3-10% tBA resulted in very fine needle-shaped ice crystal
- 1^{o} drying time in the presence of tBA << without tBA
- There was no formation of mass-transfer hindrance skin in the presence of tBA
- Dry product resistance without tBA = 10 cm^2 torr hr/gm

  Dry product resistance with tBA = 0.5-3 cm^2 torr hr/gm
- Specific surface area of freeze dried cake of 5% sucrose in the presence of 5% tBA = 8.57 m^2/gm

  Specific surface area of freeze dried cake of 5% sucrose in the absence of 5% tBA = 0.67 m^2/gm
- Addition of tBA did not change collapse temperature, but the rapid rate of sublimation prevented the product from reaching the collapse temperature

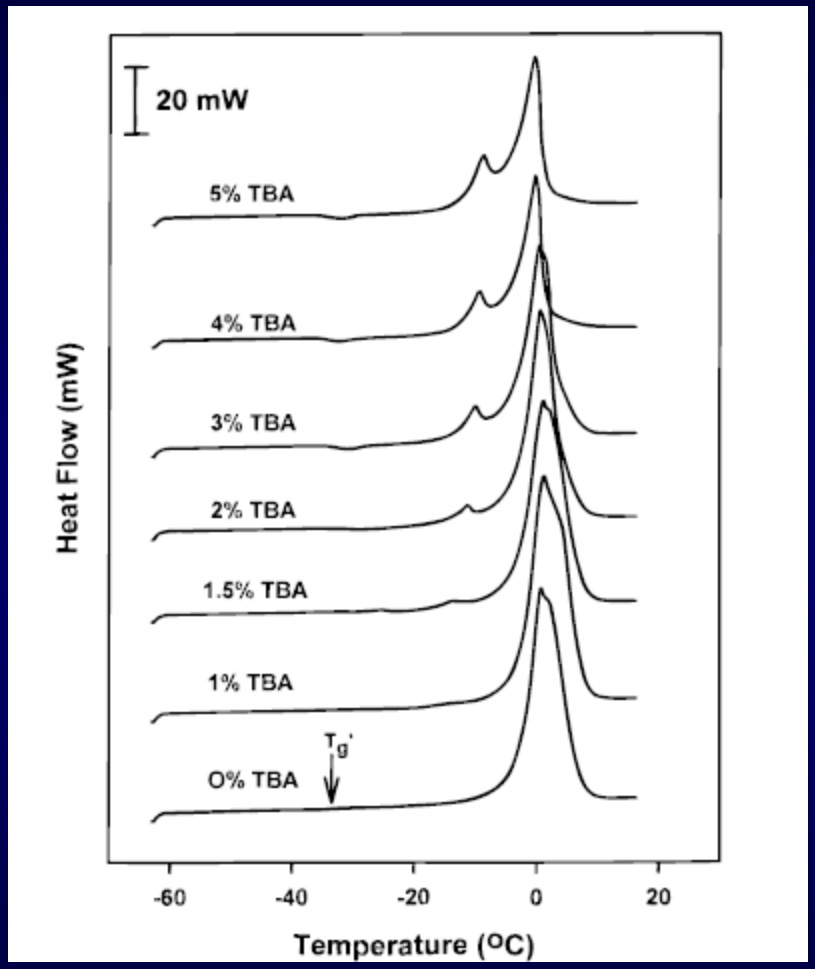
Freeze drying of t-BA/water/solute

- Thermal behavior
- Effect of physical state of the solute
- Effect of freezing rate
- Effect of secondary drying time and temperature
- Effect of cake thickness

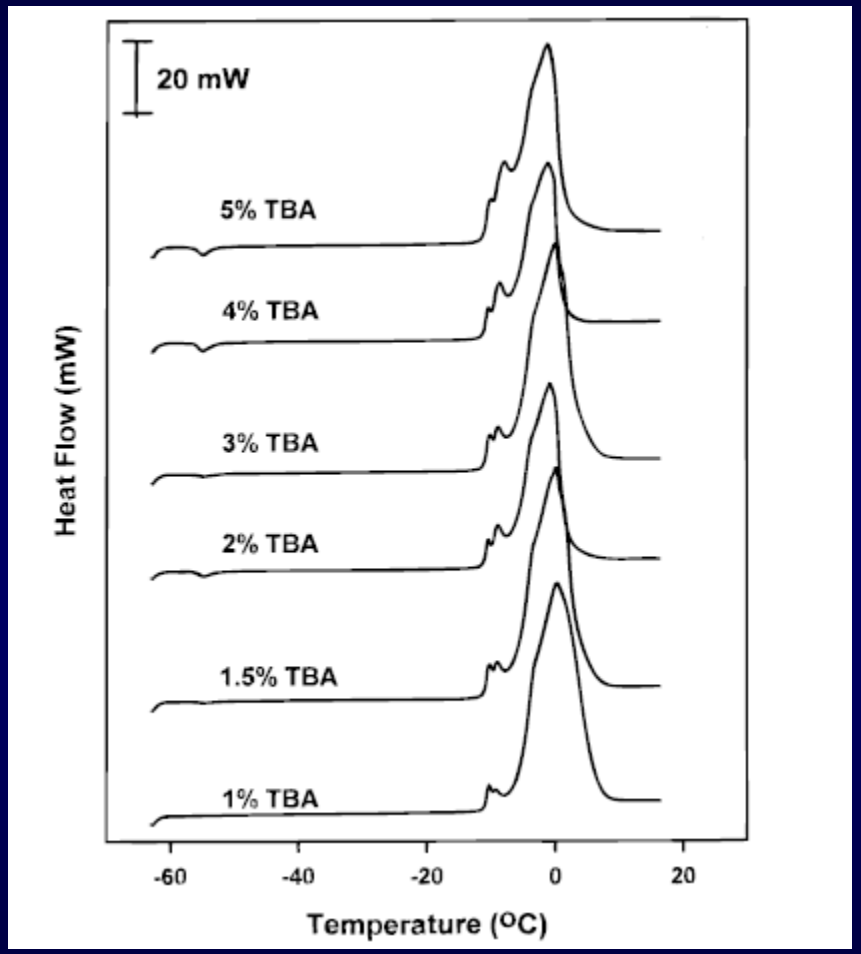
Wittaya-Areekul and Nail (JPS, 1998)

Thermal behavior

Sucrose 5%



Glycine 5%

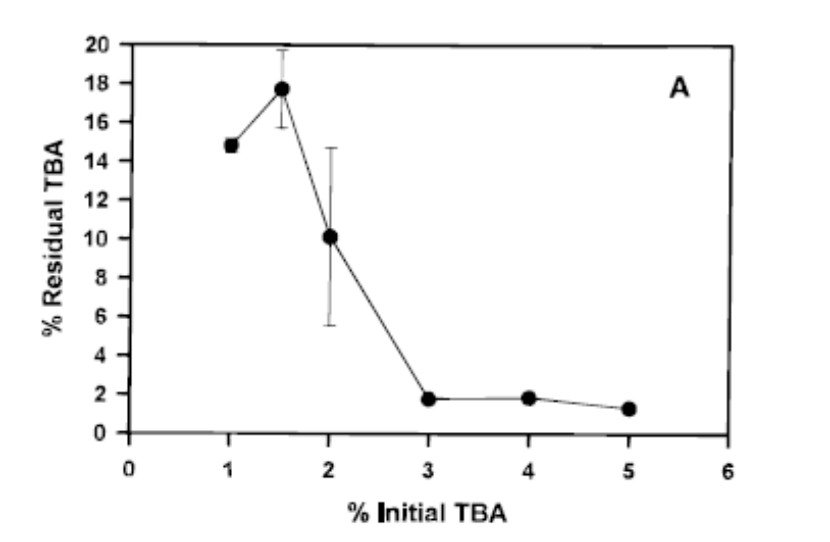


DSC thermograms of solutions containing Sucrose and Glycine along with tBA

Wittaya-Areekul and Nail (JPS, 1998)

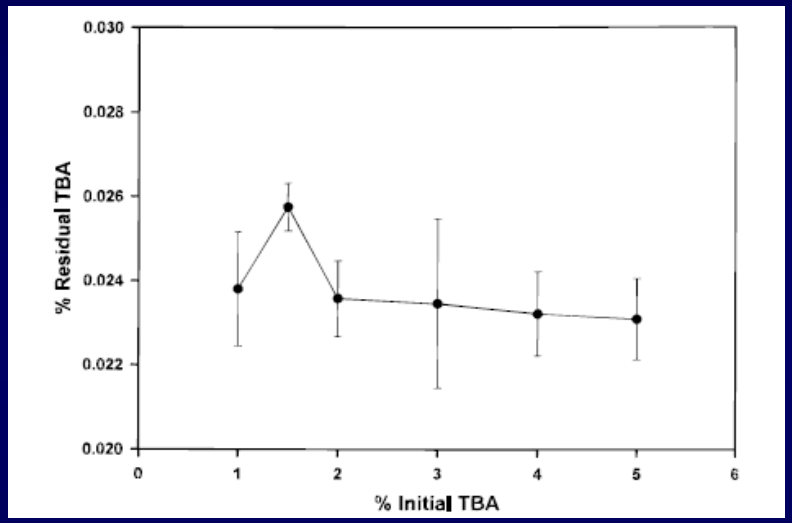
Effect of physical state of the solute

Sucrose 5%



- **Below 2% threshold, tBA remains dispersed in the amorphous phase, above 2%, remains as part of eutectic system**
- **Above 2%, residual tBA drops dramatically**

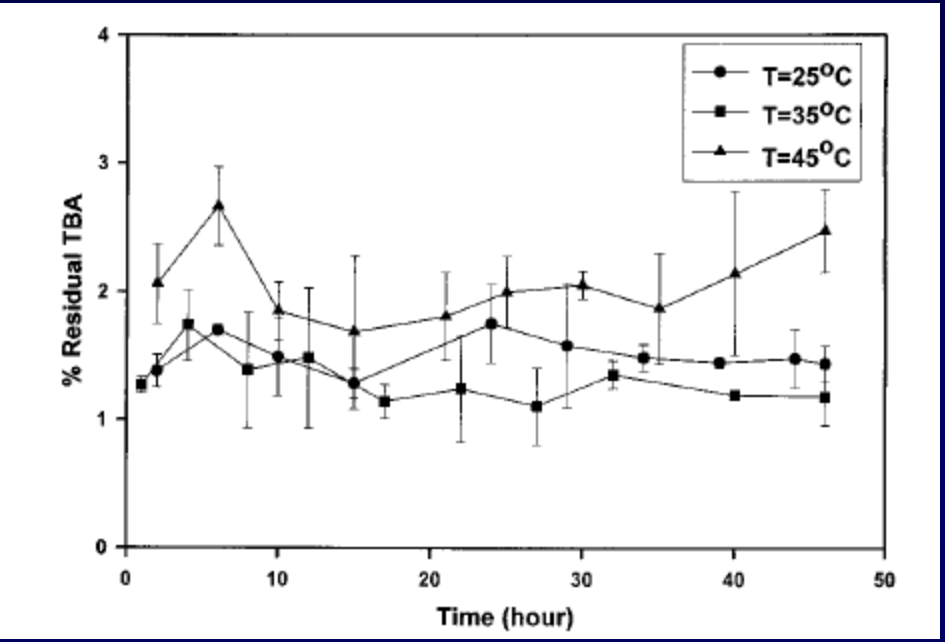
Glycine 5%



- **tBA remains crystallized irrespective of concentration**
- **Residual tBA levels much lower**
- **Residual tBA is limited to portion in amorphous phase**

Wittaya-Areekul and Nail (JPS, 1998)

Effect of secondary drying time and temperature



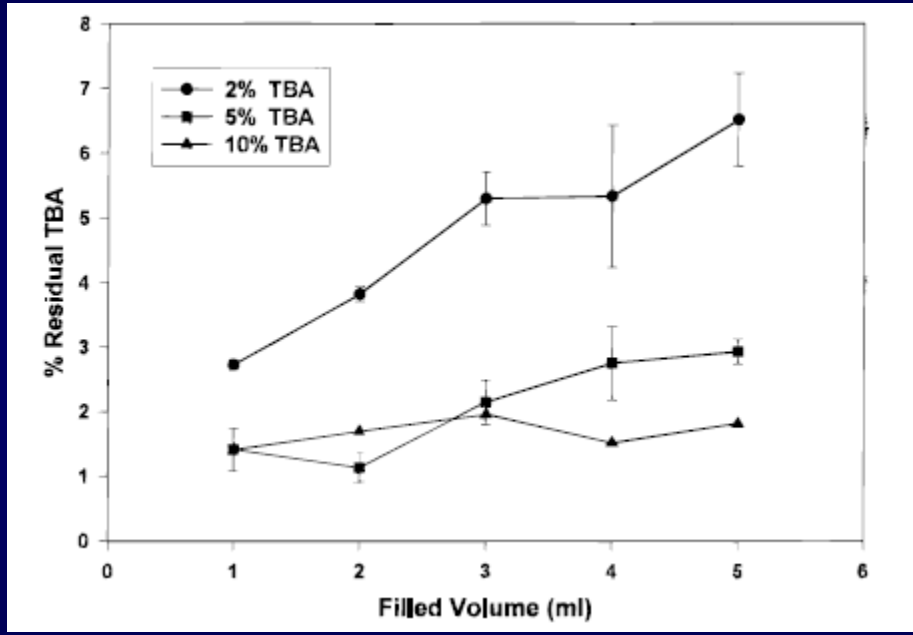
Residual tBA as a function of time during 2º drying

- **tBA plateau level is flatter**
- **Only slightly affected by shelf temp in the range 25-45 C**

Levels approach plateau
Plateau level decreases shapely with temp
Rate increases as product surface area increases
Chamber pressure and thickness : No effect

Wittaya-Areekul and Nail (JPS, 1998)

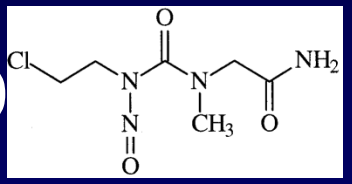
Effect of cake thickness on residual tBA (5% sucrose)



- **In 2% level, tBA did not freeze, and the rate limiting step in the 2º drying is either diffusion controlled or evaporation though the cake**

Wittaya-Areekul and Nail (JPS, 1998)

Example: Use of tBA for water-unstable compound



- Water unstable compound – SarcNu (a nitrosourea)
- Freeze dried in 100% tBA (5 mg/mL)
- Needle-shaped loose crystals upon freezing
- Drying was 2.5 times faster than in water
- Residual tBA level <0.001%
- Stable cake for long period
- Factors for success:
  - Crystal morphology, high volatility, high viscosity, low reactivity

Nina Ni et al., IJP, 2001

Use of tBA for water-insoluble compound

- Many examples of cytotoxic compounds which are insoluble (< 0.1 mg/mL) and unstable (long-term) in commonly acceptable injectable solvents/excipients
- Addition of tBA increases solubility significantly
- Typical level of tBA ranges 5-50%
- The dried cake can be reconstituted in physiologically acceptable diluents

Conclusions

- Addition of tBA in the solution for lyophilization affects the drying rate by factors such as:
  - Providing a component that dries faster than water
  - Modifying freezing pattern that leads to increased porosity, high SSA, and thus less resistant dry layer
  - Maintaining the product temperature below collapse even at high shelf temperatures
- Lower residual tBA in the final products are seen when it crystallizes out in the matrix
- tBA is an useful co-solvent for freeze drying of injectables