Development of a Rapid and Efficient Lyophilization Process

Indu Javeri, and K. Nellaiappan
CuriRx, Inc, 205 Lowell St, Wilmington, MA 01887, Email: ljaveri@curirx.com

Abstract: Lyophilization, also known as freeze-drying, is a widely used method to stabilize various pharmaceutical drugs including small molecules, peptides, proteins, and oligonucleotides. Lyophilization is a complex and expensive process. The development of process parameters is dependent on the determination of various factors like temperature, pressure, freezing, glass transition, etc. The entire process of freeze-drying may require up to 240 hours depending upon the formulation. Traditionally there are three steps in the freeze-drying process: freezing, primary drying, and secondary drying. We have developed a two-step process that requires minimum optimization and produces a lyophilized product equivalent to the one produced with a conventional three-step cycle. We have successfully used this rapid lyophilization cycle for several candidate molecules including a therapeutic antibody. Data presented here clearly show that this alternative rapid and efficient lyophilization process is widely applicable, requires minimal effort to optimize, and requires less time to freeze dry, making the lyophilization process easily scalable and more cost-effective. The equivalency of the lyophilized product utilizing these two methods of freezing drying process was determined by analysis of cake appearance, cake structure as seen by SEM, reconstitution time, and product-specific accelerated stability evaluation.

Introduction

Over the past several decades, lyophilization/freeze-drying has been used to produce stable drug products with a much-improved shelf-life. Lyophilization is a process of arresting water-mediated catalytic reactions. The conventional process involves three steps: freezing the product at or below -40°C for several hours, primary drying (where shelf temperature is raised such that the product temperature remains at or below glass transition –Tg') which removes most of the water by sublimation' and secondary drying where shelf temperature is further raised to an ambient or higher temperature that removes water by desorption. This three-step conventional lyophilization cycle involves significant time in optimizing the lyophilization cycle and each cycle may run for several days. We have developed a two-step process that involves freezing and drying at a sub-ambient temperature under low pressure (<100mTorr). Several therapeutic molecules have been lyophilized using a rapid two-step lyophilization cycle and compared with the conventional lyophilization cycle. Data presented here indicate that both processes show equivalent lyophilized products using several techniques commonly used in the pharmaceutical field.

Methods

Formulation: Humanized mAB (20 mg/mL in 10mM Succinate, pH 5.5, 6% Sucrose, and 0.02% polysorbate-20. The fill configuration is 3mL fill in 10 mL type 1 clear glass vial, 20mM single vent flurotec stoppers. The batch size is 50 vials.

Lyophilization Cycle: Parameters

Conventional Lyophilization Cycle

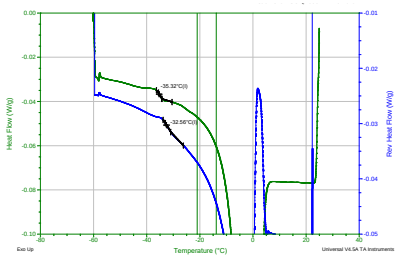
Temp (ºC)	Ramp rate (ºC/min)	Hold Time (min)	Total Time (min)	Pressure (mTorr)
5	0.5	60	60	
-50	0.3	90	270	
-10	0.2	880	1120	100
20	0.15	150	270	120
			1720	

Rapid Lyophilization Cycle

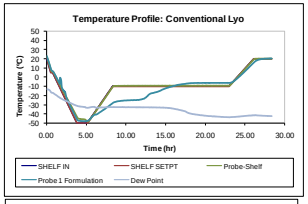
Temp (ºC)	Ramp rate (ºC/min)	Hold Time (min)	Total Time (min)	Pressure (mTorr)
5	0.5	15	15	
-50	1	90	150	
20	1	630	700	100
			865	

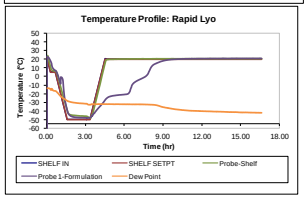
Results

MDSC analysis to evaluate Tg'



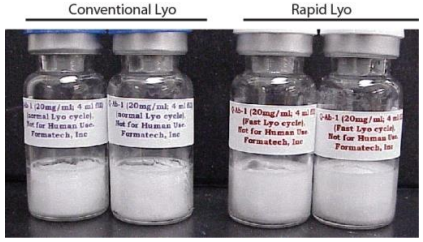
Results (contd.)





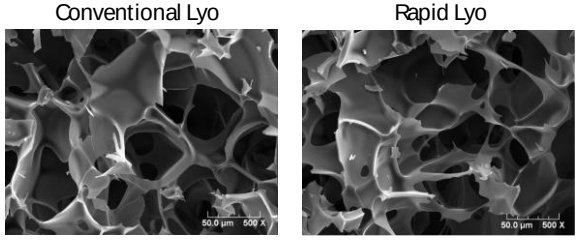
A rational approach will reduce the lyo cycle time, money, and product stability

Lyophilized product



Microscopic cake structure

SEM photographs of the cakes (middle section)



Results (contd.)

Moisture content and Reconstitution

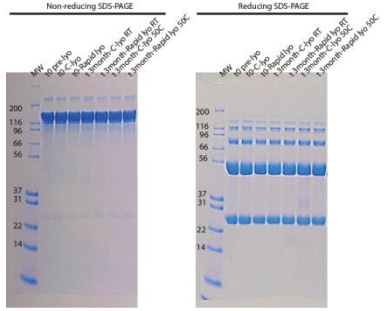
Storage	Mositure Content (%)		Reconsitution time	
	Conventional lyo	Rapid Lyo	Conventional lyo	Rapid Lyo
t0	2.56		25 sec	
3 mo RT	2.43	2.53	24 sec	22 sec
3 mo 50°C	2.17	1.85	23 sec	25 sec

Accelerated Stability

Right angle light scattering (RALS) and SEC-HPLC

	RALS (450)		% Purity (SEC)		% Aggregation (SEC)	
	Conventional Lyo	Rapid Lyo	Conventional Lyo	Rapid Lyo	Conventional Lyo	Rapid Lyo
t0 pre-lyo	2.9		96.19		3.32	
post lyo t0	2.66	2.56	95.96	96.26	3.47	3.25
3mo RT	2.19	2.21	96.24	96.17	3.05	3.09
3mo 50℃	2.19	2.19	95.53	95.4	3.69	3.89

SDS-PAGE



Key Conclusions:

1. We have developed a rapid and efficient lyophilization cycle that requires minimum optimization
2. Accelerated stability data indicate that this process is equivalent to the conventional lyophilization cycle
3. This process has been applied to several therapeutic molecules including antibodies and small molecules, reducing the lyo cycle time (in some cases, reducing the time from 87 hrs to 27 hrs), money, and keeping the stability
4. This process is scalable to cGMP production scale lyophilizers