## Materials List for:

## Ammonia Fiber Expansion (AFEX) Pretreatment of Lignocellulosic Biomass

Shishir P. S. Chundawat<sup>1</sup>, Ramendra Pal<sup>1</sup>, Chao Zhao<sup>1</sup>, Timothy Campbell<sup>2</sup>, Farzaneh Teymouri<sup>2</sup>, Josh Videto<sup>2</sup>, Chandra Nielson<sup>2</sup>, Bradley Wieferich<sup>3</sup>, Leonardo Sousa<sup>3</sup>, Bruce E. Dale<sup>3</sup>, Venkatesh Balan<sup>4</sup>, Sarvada Chipkar<sup>5</sup>, Jacob Aguado<sup>5</sup>, Emily Burke<sup>5</sup>, Rebecca G. Ong<sup>5</sup>

Correspondence to: Shishir P. S. Chundawat at shishir.chundawat@rutgers.edu, Venkatesh Balan at vbalan@uh.edu, Rebecca G. Ong at rgong1@mtu.edu

URL: https://www.jove.com/video/57488

DOI: doi:10.3791/57488

## **Materials**

Name	Company	Catalog Number	Comments
Safety Equipment/PPE			
Ammonia Monitor	CanarySense	BW GAXT-A-DL	Single gas detector, Ammonia (NH3), 0 to 100 ppm
Cryogenic gloves	Amazon	B01L8WA238/B01L8WA1H0/ B01L8WA1O8	Keep hands protected when handling liquid ammonia
Ear muffs	3M	Н7А	Ear muffs to protect hearing when releasing ammonia at end of pretreatment
Face shield	-	-	Wear while handling ammonia
Heat protective gloves	Grainger	2EWX1/2EWX2/2EWX3	Showa heat resistant gloves, max temperature 500°F
Nitrile gloves	-	-	Wear while mixing biomass to prevent contamination
Reagents			
Anhydrous Ammonia Compressed Gas Cylinder	-	-	An anhydrous ammonia compressed gas cylinder with a dip tube is required for this process. The dip tube is essential in order to withdraw liquid ammonia from the cylinder.
Distilled water	-	-	Used to add water to the biomass to achieve the desired water loading
Milled or Chopped Corn Stover	-	-	Corn stover is not readily commercially available. Contact local farmers or agricultural extension if you wish to locate some.
Nitrogen Compressed Gas Cylinder	-	-	
Equipment			
Ammonia Cylinder Adapter	-	-	CGA fitting that depends on the gas cylinder. Matheson is a good source. Some require teflon gaskets. This connects the cylinder to the ammonia delivery system. A regulator is not necessary as the system uses liquid ammonia.

<sup>&</sup>lt;sup>1</sup>Department of Chemical and Biochemical Engineering, Rutgers-State University of New Jersey

<sup>&</sup>lt;sup>2</sup>Michigan Biotechnology Institute (MBI)

<sup>&</sup>lt;sup>3</sup>Department of Chemical Engineering and Materials Science, Michigan State University

<sup>&</sup>lt;sup>4</sup>Department of Engineering Technology, Biotechnology Division, School of Technology, University of Houston

<sup>&</sup>lt;sup>5</sup>Department of Chemical Engineering, Michigan Technological University

Ammonia Delivery System (Figure 4)	Swagelok	Misc.	Stainless steel pressure cylinder and components, valves, check valves, and gauges were used for all lines potentially in contact with ammonia.
Analytical Balance	Sartorius	CPA4202S	Balance used for preparing biomass and weighing the reactors. Toploading balance, 4200g x 0.01g
Chemraz O-rings	Harvard Apparatus	5013091	Ammonia-resistant o-rings for the SS syringe
Custom Tubular Reactors (Figure 3)	Parts were purchased from McMaster-Carr, Swagelok, Omega, and Motion Industries (Dixon Fittings)	Misc.	To be compatible with ammonia, the custom reactor was constructed from stainless steel components (sanitary tube and fittings, compression fittings, quick connect, pressure gauge, thermocouple), and teflon gaskets. The maximum pressure rating of the vessel is 1500 psig, which is the maximum pressure rating of the bolted sanitary clamps.
Drying Box	-	-	Optional: an enclosed system for drying is necessary if planning to do microbial experiments to avoid contamination. Avoid drying at elevated temperatures.
High Pressure Syringe Pump	Harvard Apparatus	70-3311	Infuse/Withrdraw PHD ULTRA HPSI Programmable Syringe Pump for transferring liquid ammonia
Moisture Analyzer	Sartorius	MA35	Moisture analyzer for determining moisture content of biomass prior to pretreatment.
Nitrogen Delivery	Misc.	Misc.	Nitrogen compressed gas cylinder, inert gas regulator (at least 1000 psig max pressure rating), lines, and valves.
Ratchet wrench and 7/8" socket	-	-	Ratchet and socket to quickly tighten and open bolts on the sanitary clamp. Can be purchased anywhere.
Retractable Thermocouple Cables	Omega	RSC-K-3-4-5	Retractable thermocouple cable. You need one for each reactor.
Stainless Steel Syringe	Harvard Apparatus	702261	Stainless steel syringe for tranferring ammonia to the reactors.
Temperature Monitor	Omega	HH12B	Dual input temperature monitor. You need one for every two reactors.
Voltage Controller	McMaster-Carr	6994K11	Variable-Voltage Transformer for controlling heating to the reactors. You need one for each reactor.
Supplies			
Metal Scoops, Spoons and/or Spatulas	-	-	For transferring biomass for weighing, mixing, transferring into the reactor and removing from the reactor at the end of the run
Plastic Bowls or Tubs	-	-	Used for mixing the biomass with the water. Any bowl or tub could be used.

Spray Bottle	-	-	Used to add water to the biomass to achieve the desired water loading
Wide-Mouth Funnel	-	-	Any funnel that has a bottom opening 0.5-1.0 inches diameter.
Wooden Dowel	-	-	1-1.5" diameter wooden dowel to assist with loading/unloading the reactor
Consumables			
Glass Wool	Sigma-Aldrich	CLS3950-454G	For packing the top of the reactor to prevent biomass escape and clogging the tubing
Plastic Press-to-Close Bags	McMaster-Carr	1959T24	Bags for storing processed samples and for transferring to drying box
Plastic Tote	-	-	Used to transfer pretreated biomass to an alternate location for drying
Plastic Weighboats or Metal Trays	-	-	Used to catch the biomass when removing from the reactors, and for storing the samples while drying