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Freeze Drying Plant and Food Samples

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Protocol status: Working
We use this protocol and it's working

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Abstract

This protocol describes a standardized method for freeze-drying plant-based food samples and is optimized to preserve structural integrity, nutritional content, and bioactive compounds. Samples are first gradually frozen to form large ice crystals, then subjected to controlled primary drying under vacuum, followed by secondary drying to remove bound moisture. By carefully managing sublimation and temperature profiles, this method minimizes collapse and volume loss, while maximizing retention of delicate nutrients and phytochemicals. The resulting lyophilized materials are ideal for long-term storage, downstream biochemical analyses, or other studies. This approach balances process efficiency with sample quality, making it broadly applicable for research in food science, nutrition, and plant biochemistry.

Materials

Harvest Right Freeze-Dryer Mk 10.967a

Troubleshooting

Problem

Dark spots or signs of residual moisture

Solution

If dark spots or other signs of residual moisture are present, the system may have been overloaded. Re-freeze samples immediately and start the process over the following day. Depending on the amount of remaining moisture, the samples may only need to be freeze dried for an additional 48-72 hours

Preparation

14h 10m

1 Prior to freeze drying, create and save the freeze drying profile (details are found in "run profile" section) 10m

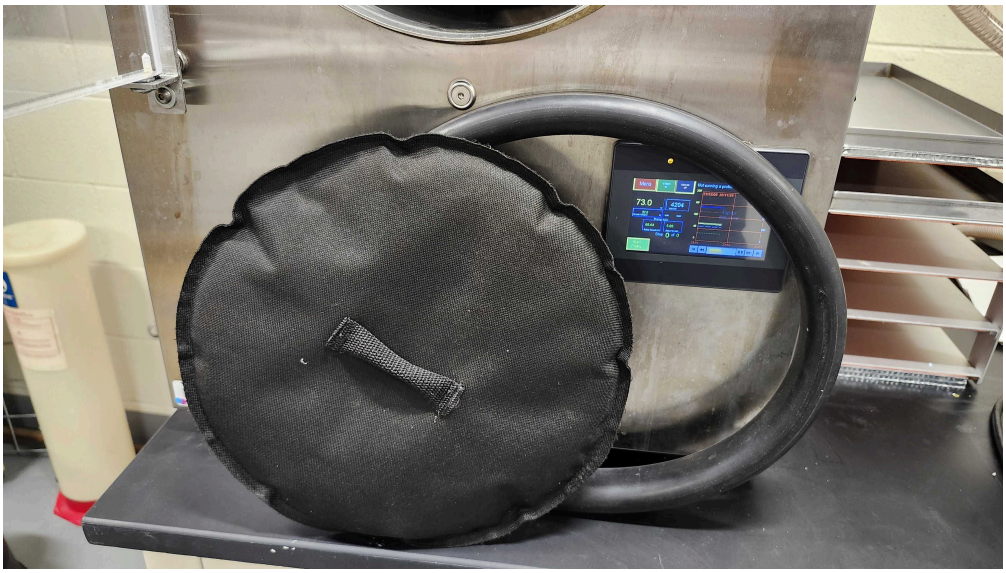
2 Ensure samples are frozen (ex: leave samples in a -20 °C freezer for at least one night) 10h

2.1 Freezing samples more slowly will result in relatively larger ice crystals making lyophilization more efficient

3 Power on freeze dryer (Harvest Right Freeze Dryer Mk 10.967a in this protocol, but these parameters will likely work on other systems) and set it to chill to -30 °F 3h

3.1 The freeze dryer will take ~3 hours to reach temperature

3.2 gTo chill more efficiently, install the insulating cover and gasket, close the door, and close the drain hose



Insulating cover and gasket

Closed freeze dryer with insulating cover and gasket installed

Drainage hose in the closed position

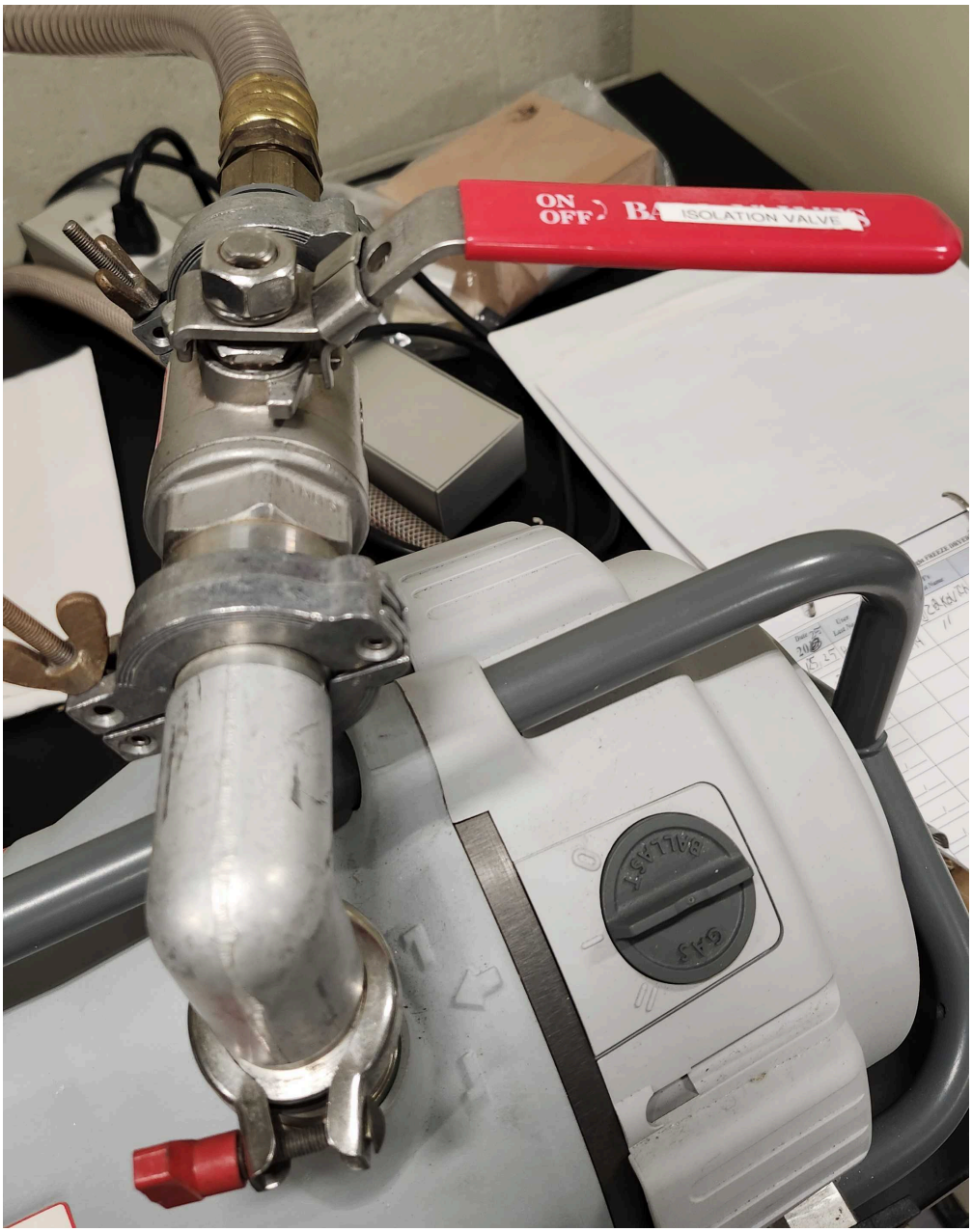
4 Move your samples to a walk-in refrigerator. There, uncap all your samples and cover each sample tube opening with a piece of cheese cloth 20m

Example of tubes covered with cheese cloth

4.1 Secure the cheese cloth in place with a rubber band 5m

5 Move the samples back to a freezer. Allow the samples enough time to become frozen again (typically at least 20 minutes in a -80 °C freezer) 20m

6 Once the freeze dryer reaches a temperature of -30 °F, close the vacuum pump line and start the vacuum pump. 15m



Vacuum pump in the closed position

6.1 Allow the vacuum pump to warm up for 15 minutes before loading your samples.

Loading samples

9m

7 Remove your samples from your freezer and then place them inside the freeze dryer on the shelf. Replace the insulating cover and latch the freeze dryer door closed

3m

7.1 Make sure you place the sample tube caps on the lower freeze dryer shelf. They may have a bit of sample material or residual moisture on them that also need to be freeze dried. Not freeze-drying the caps will alter total weight and moisture content estimates.

8 Set the freeze dryer to have a maximum pressure of 700 mTorr and a minimum pressure of 300 mTorr 1m

Freeze dryer screen. The pressure range is set to 300 (Low) and 700 (High)

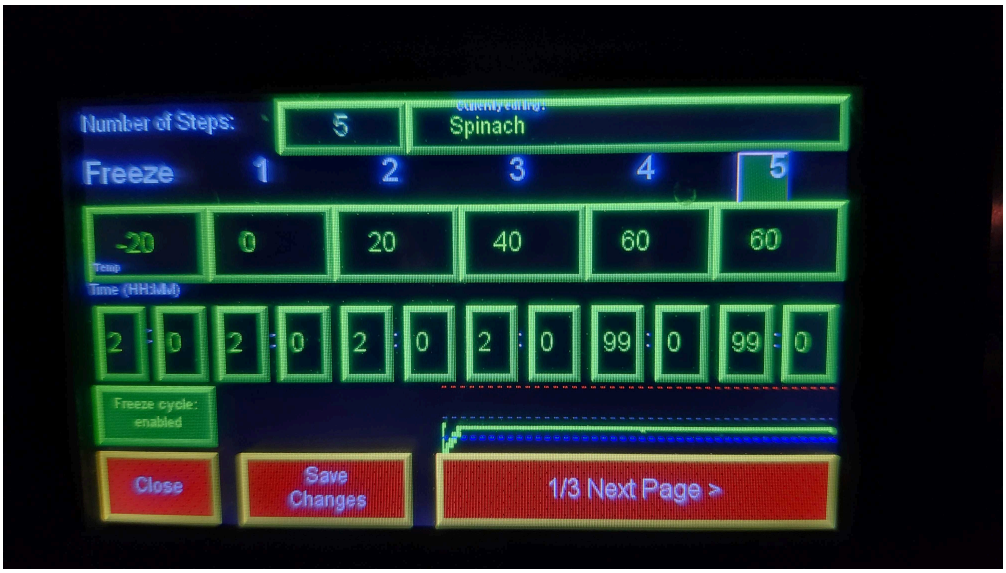
9 Ensure the drainage hose is closed. Then, open the vacuum pump line slowly to allow the pressure to decrease (~1 minute) 1m

Vacuum pump in the partially open

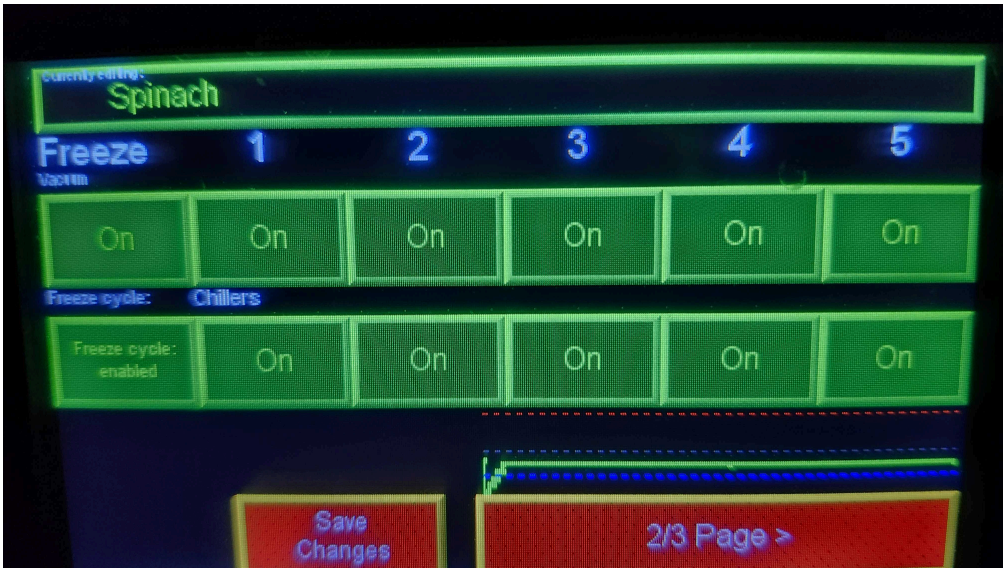
10 Wait until the pressure reaches your pressure range (300-700 mTorr) then proceed to next section 4m

Run Profile 5d 0h 2m

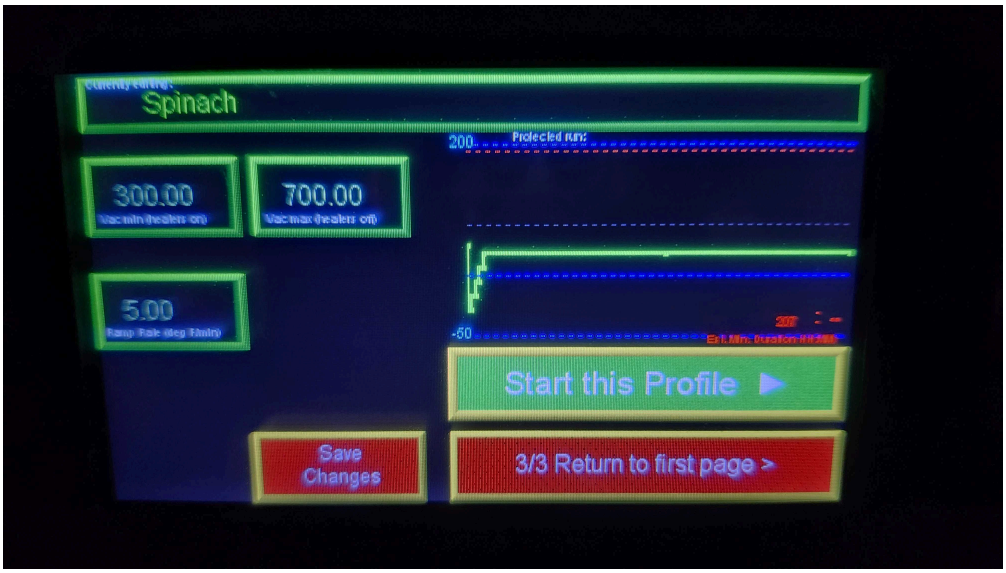
11 Start the profile to begin freeze drying. The profile needs to have a ramp rate of 5 °F per minute and must consist of the following steps and settings: 2m



Freeze dry protocol steps



"Vacuum" and "Chiller" should be activated for all steps



Pressure should range from 300 to 700 and the ramp rate (°F/min) should be 5

11.1 step 0: temperature set to -20 °F for 2 hours

11.2 step 1: temperature set to 0 °F for 2 hours

11.3 step 2: temperature set to 20 °F for 2 hours

11.4 step 3: temperature set to 40 °F for 2 hours

11.5 step 4: temperature set to 60 °F for 99 hours

11.6 step 5: temperature set to 60 °F for 99 hours

12 Depending on how much total water content is in your samples, leave the samples in the freeze dryer anywhere from one day to five days 5d

12.1 The more total water content your samples are, the longer you should leave the samples in the freeze dryer. Five days is recommended for most batches of ~200-250 g of total plant material

Unloading freeze dryer

10m

13 Close the vacuum pump line, then end the freeze dryer profile 2m

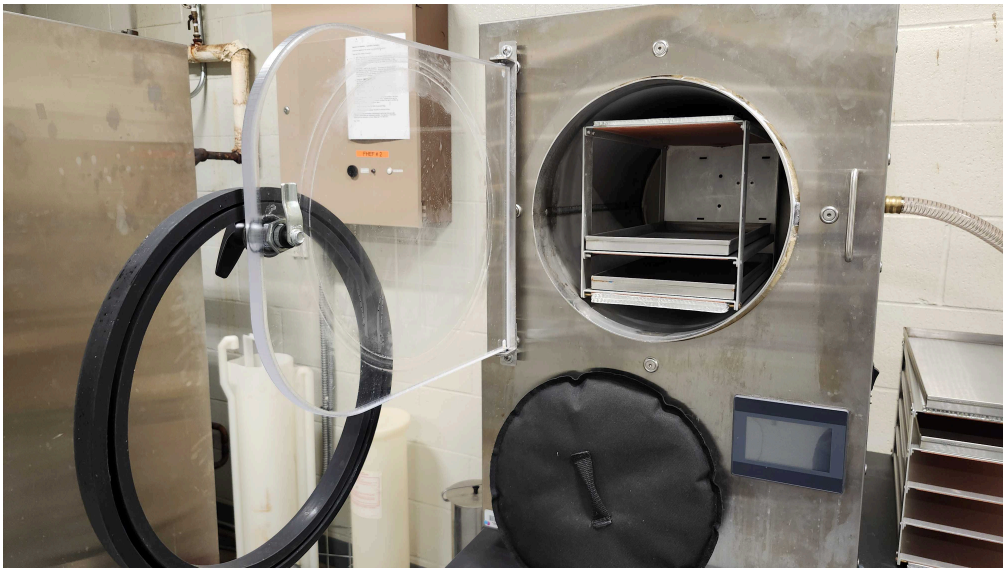
13.1 Make sure you close the vacuum pump first to avoid rapidly pressurizing the samples

13.2 Ensure that vacuum pump is powered off once isolation valve is in the closed position

14 Slowly open the drainage hose (~1 minute) to allow the pressure in the freeze dryer to equalize 1m

15 Remove samples from the freeze dryer shelf. Immediately remove the cheese cloth and reinstall the sample caps 3m

16 Turn off the freeze dryer completely. Leave the door open and keep the insulating cover removed to allow the the inside to defrost and dry out 2m



Freeze dryer post shutdown

17 Weigh capped samples if necessary and move samples to their storage destination. For some applications, room temperature can be suitable, but do sufficient testing to ensure any compounds of interest are stable over time. 2m