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© Using the Thermo SPD1010 speedvac concentrator centrifuge for drying down peptides for LC-MS analysis

In 1 collection

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ABSTRACT

This protocol details the procedure of using the Thermo SPD1010 speed vacuum concentrator centrifuge to dry peptide samples for storage and submission for LC-MS analysis. Peptides may be stored at § 4 °C or § -20 °C when dried down this way.

ATTACHMENTS

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COLLECTIONS (i)

Researcher led sample preparation for LC-MS using the BioMS research core facility

KEYWORDS

Thermo spd1010, Vacuum concentrator centrifuge, Speedvac



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Researcher led sample preparation for LC-MS using the BioMS research core facility

GUIDELINES

Initial assumptions:

- You have peptides in non-chlorinated solvent that you need to remove, and thereby concentrate and store your sample.
- The speed vacuum (speedvac) process works by drying the sample at the bottom of the vial recovery and storage. It takes place under vacuum conditions, which promotes solvent evaporation in the speedvac chamber. Centrifugation generates a centrifugal force to prevent bumping (bubbling), boiling, and physical loss of the sample. Samples are maintained in liquid state at sub-ambient temperature throughout the concentration process, preventing loss of activity or damage to heat sensitive substances. By applying a controlled vacuum the boiling point of the solvents can be lowered to the point that the liquid vaporizes with minimal heat which prevents oxidation of samples during the drying process.
- For 120 to 150 uL of peptides in approximately 30% acetonitrile, allow 75 to 90 minutes to dry the peptides and evaporate the solvent.

MATERIALS TEXT

Screw-top caps for Waters LC-MS vials with the PTFE/Silicone septa removed.



Caps used in drying down process

SAFETY WARNINGS

Try to minimise contact with Cryocool - use appropriate safety wear.

BEFORE STARTING

Locate the Thermo SPD1010 speedvac, it is at corner of the sample prep lab in room B2075.



Locate the following consumables:

Vial caps with the white septa removed (contained in a blue box)

We will ensure that there is sufficient Cryocool present before use.

A line on the wall of the stainless steel trap indicates the minimum appropriate level. Cryocool conducts heat away from the glass condensation flask, allowing vapours to condensate on the flask walls.

Before you start: On the right hand side of the machine, there is a white rubber gasket - It is important to ensure that the white flask insulating seal is over the glass flask to secure the flask in the cooling chamber.

Speedvac drying: 45m

Turn the power switch located on the rear right hand side of the unit to the ON position. Wait © **00:45:00** before starting drying.

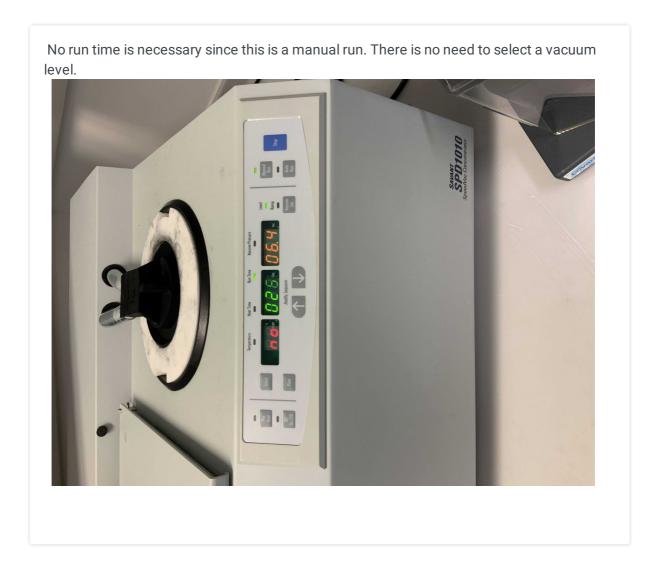
This is to allow the system to come to temperature. The cover lock disengages, allowing the top cover on the left side of the unit to be opened.



After waiting for the system to come to temperature, add the vials to the centrifuge by placing the neck of the vial in and under the hole, and secure into place by screwing a blue vial cap onto it.

Do this for all of your samples, and balance them as you would for any centrifuge, across the spindle.

- 3 Close the lid, and switch on the vacuum pump on top of the cold trap.
- 4 Use a manual run for drying, this allows you to intermittently check the progress of the vacuum drying procedure.
- 5 Use the SELECT button and the up/down keys, set the temperature to "NO" for no heat. Set the heat time to "CCC".



6 Press the "MANUAL RUN" button. The cover locks and the rotor starts turning. The decimal point blinks. If the cover is not closed, the display will show "Lid" and the run will not start.

7	Press the "STOP" button to end the manual run. The display will show "END", the valves Will 5m click, and the chamber will isolate from the vacuum pump allowing air into the chamber.
8	After the rotor stops spinning, the cover lock disengages and the display reverts to the last set parameters, and the unit will emit several audible beeps.
9	Open the cover and remove the samples.
10	When the lid click open, lift it and remove the vials, if dry, you can replace the speedvac cap with ordinary blue LCMS caps, located on the workbench in a square box.
11	Switch off the vacuum pump If no one else has samples in the speedvac.
12	Proceed to sample submission.