

Retsch Grinding and Homogenization Procedure

Pipet 250 µl of the 60% acetonitrile master-mix into each screw top 1.5-ml mi

For each person working, transfer one frozen sample from the 1.5 ml snap cap
Clean scissors with nanopure water and dry. Keep the rest of the samples in li

Working quickly for each sample and keeping the snipped samples on ice, rep

Retsch:

The Teflon tube holders should be placed in the dry ice to chill during the sam

Load microfuge tubes with samples into the two tube holders in equal numbe:

NOTE: Make sure the screw cap tubes are tightened very tight to prevent leaks.

Run one cycle of 2 minutes at the maximum (30 Hz) setting for Cancer slices. I

Remove the tubes to the ice tray and place the holders in the dry ice.

Transfer the mixture to the 15ml tube. Wash the 1.5 ml tube and balls with 50

microfuge tubes (up to 10 can be processed together). Tubes **MUST** be balanced

microfuge to the new 1.5 ml screw cap microfuge containing the 60% acetonitrile
q N2 until ready for 60% acetonitrile addition.

eat snipping of remaining frozen samples (up to 10). Each person should not touch

sample snipping process.

rs and with symmetry in position. Place the two loaded tube holders in the Ret

Run two more cycles for Normal slices (total 6 minutes, depending on how big

10 μ l 60% acetonitrile mixture 3 times. Transfer the mixture to 15 ml tube each

in the left and right Teflon tube holders, so an even total number of tubes must be used. Immediately snip the slice with the microscissors. Rinse the microscissors with acetonitrile. Do not use the microscissors to do more than 5 samples at a time.

Place the tubes in the holders with the tops facing one another. Fasten securely.

Grind the slice in the holder (adjust grinding time to achieve complete homogenization).

Grind for 10 minutes. Add additional 60% acetonitrile to a total of 3.5ml volume.

t be prepared.

rs with 250 μ l of the 60% acetonitrile master-mix into the sample tube. Add or

re set of 6 beads to the tube.