Precellys Homogenization and Quenching Procedure

For every tissue sample we use:
2 ml 100 % acetonitrile
1.3 ml dH_2O
0.2 ml 0.2 mM Tris-Cl pH8.0 (40 nmoles of Tris)
Total volume = 3.5 ml

To avoid mixing up 60%AcN with Tris and 60% AcN without Tris, my suggestion is t

We can make master mix for 20 samples as follows:
Master Mix:
2 ml x 20 = 40 ml 100% acetonitrile
1.3 ml x 20 = 26 ml dH2O
0.2 ml x 20 = 4 ml 0.2 mM Tris-Cl pH 8.0 (40 nmoles Tris)
Total V = 70 ml

Slice Pre-treatment:

* Add 100 ml master mix to snap-cap tube (depending on the slice size and shape
* Use microscissors to cut the slices inside the snap-cap tube
* Use pestle to smash the pieces against the tube wall (BE VERY CAREFUL)
* Use microscissors again to cut the slices inside the snap-cap tube
* Use pestle again to smash the pieces against the tube wall (BE VERY CAREFUL)
* Wash the pestle with 100 ml master mix
* Wash the scissors with 100 ml master mix
* Transfer sample to Precellys tube containing beads using 1 ml pipet tips cut at th
* Wash snap-cap tube and pipet tip with 2 x 100 ml master mix. KEEP the tip.

In this way, about 500 ml of pre-cut slices will be further homogenized using Prece
Precellys:
Add ice to coil and wait till air T goes down to 16 ^{0}C.
Use 3 cycles of 5000 rpm - 5 seconds interval in between runs.
At this point, you have 3 ml of master mix to use for transferring the homogenized
Rinse the beads and tube with 3 x 1.0 ml of master mix, transferring to the 15 ml t

to prepare a master mix with all 3 components described above.

you may need to use another 50 or 100 ml - if this is the case, remember that the t

ıe tip end with razor blade to facilitate transfer to small tissue chunks.

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d/quenched sample to 15 ml conical tube and washing the tube with beads and the
tube.

final volume in the Precellys tube with beads should be around 500 ul)

e pipet tip you saved before.