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Zip-Tip purification

Guidelines for sample preparation
(How to protect your samples from contamination with keratin)

- **Clean your bench**
- **Try to avoid contact of samples and solutions with dust, skin or hair**
- **Wear gloves at all times**
- **All reagents should be prepared fresh**
- **Use ultra-pure water for all solutions**

SOLUTIONS

Buffer A:
98% MilliQ-H_20
2% CH3CN
0.1% TFA or FA

Buffer B:
65% CH_3CN
35% MilliQ-H_20
0.1% TFA or FA
Use HPLC-grade Acetonitrile and FA, and MilliQ-H_20.

1. Acidify sample (Vol 20-100 µl) by adding TFA (recommended) or FA (0.1 % final concentration)

2. ZipTip equilibration

   - Aspirate **Buffer B** (10 µl) into the tip. Dispense into waste. Repeat.

   - Aspirate **Buffer A** (10 µl) into the tip. Dispense into waste. Repeat.

3. Bind and Wash the peptides/proteins

   - Take 10 µl of sample. Aspirate and dispense the sample (repeat 10 x). Dispense.

   - Wash with **Buffer A** (10 µl). Dispense into waste. Repeat 4x.

4. Elution

   - Elute with 10 µl with **Buffer B** in new tube.

   - dry in vaccum centrifuge

   - resuspend in 10 µl **Buffer A.**

Or sample spotting on a MALDI

4. Elution and spotting on MALDI plate

   - Pipette 1-2 µl of matrix (alpha-cyano) in **50 % water, 50 % acetonitrile, 0.5 % TFA.**

   - Spot the sample on MALDI target. Leave for 10 minutes to dry.