SEP-PAK C18 PURIFICATION

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

- TRY TO AVOID ANY CONTACT OF SAMPLES AND SOLUTIONS WITH DUST, SKIN OR HAIR
- CLEAN YOUR BENCH
- WEAR GLOVES AT ALL TIMES
- ALL REAGENTS SHOULD BE PREPARED FRESH
- USE ULTRA PURE WATER FOR ALL SOLUTIONS

SAMPLE SUBMISSION

- PROVIDE SAMPLES IN 1.5 ML TUBES (NO SCREW CAPS)
- LABEL YOUR TUBE WITH YOUR NAME, SAMPLE NUMBER AND DATE
- PLEASE FILL IN ONLINE SAMPLE SUBMISSION FORM TO PROVIDE US WITH MORE INFORMATION ABOUT YOUR SAMPLE (PROTEIN ORIGIN, TAXONOMY, CONCENTRATION)

1. Attach a syringe to the Sep-Pak column (both should be attached to a vacuum manifold)

2. Equilibrate the column:
   – flush the column with 5 ml of solution B
   – flush the column with 10 ml of solution A

3. Add the peptide digest sample onto the column and let it into the Sep-Pak column

4. Wash the sample with 10 ml of Solution A

5. Elute with 2 x 1 ml of solution B (collect your purified sample in a 2 ml tube)

6. Dry down purified peptides completely in a speed-vac. Resuspend in Solution A (up to 20µl)

MATERIAL AND SOLUTIONS
Waters C18 Sep-Pack cartridges: Sep-Pak Plus™, WAT020515

Solution A:
98% MilliQ-H₂O
2% CH₃CN
0.1% FA

Solution B:
65% CH₃CN
35% MilliQ-H₂O
0.1% FA
Use HPLC-grade Acetonitrile and FA, and MilliQ-H₂O.