

# Preciex™ Stage-tip Instructions for:

## Peptide Desalting

### APPLICATIONS:

Complex biological samples, such as serum, plasma, urine or cell lysates frequently present a major challenge for peptide analysis because of high salt, lipid and protein content. Sample complexity reduction is an essential first step of any peptide analysis. The Preciex Desalting tips have demonstrated an excellent performance for many proteomic applications. These applications range from small protein and peptide purifications/desalting, fractionation to drug analysis. Preciex Stage tips are based on small particles densely packed and embedded in a soft membrane. They are available with several capacities and sample recovery >95%.

### Conditioning procedure

- a) Reconstitute samples in 20µl wash solvent (Soln i '98:2:0.1%'), vortex 45 sec; centrifuge 3000 g for 1 min. CHECK pH (pipette 0.5µl onto pH strip). Ensure pH is ≤3. (Adjust with 10% aqueous TFA if necessary, in 0.5µl increments, for example.)
- b) Via a pipette tip inserted in the top of the Tip, add 20µl of wetting solvent (Soln ii '80:20:0.1%') in order to wet the packing material. Centrifuge at 2000 g, it may cost 30 seconds and then visually inspect that tips are pale white.
- c) Add 20µl of wash solvent (Soln i '98:2:0.1%'), centrifuge 2000 g for 60 seconds to force the solution through the packed bed.

Suitable solutions are:

- i. Wash solvent: 98:2:0.1%, water: acetonitrile: trifluoroacetic acid (TFA)
- ii. Wetting solvent: 80:20:0.1%, acetonitrile: water: trifluoroacetic acid (TFA)
- iii. Elution solvent: 50:50:0.1%, acetonitrile: water: trifluoroacetic acid (TFA)

*Prepare fresh solvents weekly; do not pipette neat TFA with plastic pipette tips, use glass syringe. Follow protocol below for a 10µg/200µl Stage Tip, adjust solvent amounts with other Stage tip format. We do not recommend any particular buffer, because each application has its own buffer and solutions.*

### Sample Loading

Load all samples in wash solvent (Soln i '98:2:0.1%') as above, centrifuge 2000 g for 60 seconds. Ensure solvent is washed through Stage Tip, increase centrifuge time if necessary.

### Sample Washing

Wash the packed bed with 20µl volumes of the wash solvent (Soln i '98:2:0.1%'), centrifuge 2000 g for 60 seconds, repeat twice.

Place cap/Stage Tip assembly onto a new 1.5 mL Eppendorf tube; label tube w/'ST C18', sample name and your initials.

### Sample Release

Elute peptides from the packed bed with 20µl of elution solvent (Soln iii '50:50:0.1%'), centrifuge 2000 g for 60 seconds. Evaporate and reconstitute for MS analysis.

*Please be sure of that tip column is not dried out in all steps.*

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