

# Pierce<sup>®</sup> C18 Tips

**87781 87782 87783 87784**

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<b>Number</b>	<b>Description</b>
<b>87781</b>	<b>Pierce C18 Tips</b> , 10 $\mu$ L, 8 tips
<b>87782</b>	<b>Pierce C18 Tips</b> , 10 $\mu$ L, 96 tips
<b>87783</b>	<b>Pierce C18 Tips</b> , 100 $\mu$ L, 8 tips
<b>87784</b>	<b>Pierce C18 Tips</b> , 100 $\mu$ L, 96 tips

**Note:** Each tip can bind up to 8 $\mu$ g (10 $\mu$ L) or 80 $\mu$ g (100 $\mu$ L) of total peptide.

**Storage:** Upon receipt store at room temperature. Product shipped at ambient temperature.

## Introduction

The Thermo Scientific Pierce C18 Pipette Tips enable fast and efficient capture, concentration, desalting and elution of peptides. Each tip contains a monolithic C18 reversed-phase sorbent that minimizes flow resistance and provides excellent binding and recovery characteristics at a wide range of peptide concentrations. The 100 $\mu$ L tip allows processing of peptide samples for desalting after digestion and before mass spectrometric analysis. The 10 $\mu$ L tip is ideal for off-line desalting of smaller samples.

Matrix-assisted laser desorption ionization (MALDI-) and electrospray ionization (ESI-) mass spectrometry (MS) are vital tools for studying biological compounds because of the high sensitivity and mass accuracy. MS methods are commonly used for examining post-translational modifications and identifying proteins by peptide mapping; however, many buffers and compounds common to biological samples (e.g., urea, guanidine, NaCl, Tris, phosphate) interfere with both MALDI-MS and ESI-MS. Pierce C18 Tips remove interfering contaminants and release peptides in MS-compatible solutions, resulting in increased sensitivity and high-quality spectra. Although Pierce C18 Pipette Tips are designed primarily for MS applications, they may be used for applications such as peptide concentration and clean-up for peptide sequencing.

## Important Product Information

- The Pierce C18 Pipette Tips can bind up to 8 $\mu$ g or 80 $\mu$ g of total peptide in the 10 $\mu$ L or 100 $\mu$ L tip, respectively. For best results, use these tips with peptides derived from at least 20ng of protein containing at least 0.5ng of each singular peptide product. Minimum sample load requirements depend on the sensitivity limits of the downstream analysis system. Sample recovery for typical peptides is > 85%, but could be as low as 35% for hydrophilic peptides.
- To avoid inaccurate volumetric dispensing, do not use Pierce C18 Pipette Tips for measuring volume.
- For binding to C18 reversed-phase sorbents, a sample must be free of excess organic solvents such as acetonitrile (ACN) or methanol. Remove organic solvents with a centrifugal vacuum evaporator but avoid complete dryness, which might result in sample loss. Carefully dilute or resuspend sample in water with 0.1-1.0% trifluoroacetic acid (TFA) before processing with the C18 tips.
- For optimal results, prepare all solutions and collection tubes in advance and proceed with the entire procedure in a timely manner. Do not introduce air through the membrane during any portion of the procedure for optimum flow and peptide recovery.
- Plastics used during handling of peptide samples can introduce contaminants that interfere with MS analysis and result in sample loss from nonspecific adsorption. Use high-quality receiver tubes. If necessary, receiver tubes used for the final collection may be rinsed with 70% ACN/0.1% formic acid before use. Minimizing sample transfers and freeze-thaw cycles before analysis will help minimize plastic contamination and sample loss.

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## Additional Materials Required

- 10 $\mu$ L or 100 $\mu$ L pipettor
- Ultrapure water
- Acetonitrile (ACN)
- Trifluoroacetic acid (TFA)
- Autosampler vials or 0.5mL, 1.5mL microcentrifuge tubes
- Formic acid or acetic acid
- MALDI matrix (optional)
- Methanol (optional)

## Material Preparation

- Sample treatment solution: 2.5% TFA
- Wetting solution: 50:50 ACN:water; 20 $\mu$ L or 200 $\mu$ L per sample
- Equilibration solution: 0.1% TFA in water; 20 $\mu$ L or 200 $\mu$ L per sample
- Rinse solution: 0.1% TFA in 5% ACN:water; 20 $\mu$ L or 200 $\mu$ L per sample
- Elution solution: 0.1% TFA in 50-95% ACN:water for MALDI-MS or 0.1% FA in 50-75% ACN:water for ESI-MS, up to 100 $\mu$ L per sample

## Procedure for the 10 $\mu$ L C18 Tips

**Note:** For optimal flow and peptide recovery, do not introduce air through the membrane at any time during the procedure.

1. Set the pipettor to 10 $\mu$ L and secure the pipette tip tightly to the end of the pipettor for optimum tip-to-pipettor seal and sample aspiration.
2. Adjust sample to 0.1-1.0% TFA using 2.5% TFA.
3. Wet tip by aspirating 10 $\mu$ L of 50% ACN in water and then discarding solvent. Repeat once.
4. Equilibrate tip by aspirating 10 $\mu$ L of 0.1% TFA and discarding solvent. Repeat once.
5. Aspirate up to 10 $\mu$ L of sample (prepared in Step 2) into the C18 tip. For maximum efficiency, dispense and aspirate sample for 3-10 cycles.
6. Rinse the tip by aspirating 10 $\mu$ L of 0.1% TFA/5% ACN and discarding solvent. Repeat once.
7. Elute the sample as follows:

**MALDI-TOF analysis:** Slowly aspirate 2-10 $\mu$ L of 0.1% TFA in 50-95% ACN elution solution with or without matrix and dispense directly onto a MALDI plate.

**LC/MS or LC/MS/MS analysis:** Slowly aspirate 2-10 $\mu$ L of 0.1% formic acid or 0.1% acetic acid in a 50-95% ACN or methanol and dispense into an autosampler vial or well plate.

## Procedure for the 100µL C18 Tips

**Note:** For optimal flow and peptide recovery, do not introduce air through the membrane at any time during the procedure.

1. Set the pipettor to 100µL and secure the pipette tip tightly to the end of the pipettor for optimum tip-to-pipettor seal and sample aspiration.
2. Adjust sample to 0.1-1.0% TFA using 2.5% TFA.
3. Wet tip by aspirating 100µL of 50% ACN in water and then discarding solvent. Repeat once.
4. Equilibrate tip by aspirating 100µL of 0.1% TFA and discarding solvent. Repeat once.
5. Aspirate up to 100µL of sample (prepared in Step 2) into the C18 tip. For maximum efficiency, dispense and aspirate sample for 3-10 cycles.
6. Rinse the tip by aspirating 100µL of 0.1% TFA/5% ACN and discarding solvent. Repeat once.
7. Elute the sample as follows:

**MALDI-TOF analysis:** Slowly aspirate 5-100µL of 0.1% TFA in 50-95% ACN elution solution with or without matrix and dispense directly onto a MALDI plate.

**LC/MS or LC/MS/MS analysis:** Slowly aspirate 5-100µL of 0.1% formic acid or 0.1% acetic acid in a 50-95% ACN or methanol and dispense into an autosampler vial or well plate.

## Troubleshooting

Problem	Possible Cause	Solution
Poor or incomplete sample binding	High pH, lack of ion-pairing agents	Ensure TFA was added to sample
	Tip not sufficiently wetted	Check buffers and prepare new tip
	Sample contains organic solvent	Dry sample and resuspend in 10µL or 100µL of 0.1-1.0% TFA
	Sorbent became dry before adding sample	Ensure that air is not drawn into the tip and that sorbent does not dry during sample processing
	Sample not sufficiently hydrophobic to bind C18 sorbent	None
Poor or incomplete sample recovery	Highly hydrophobic sample	Use 70% ACN for elution
	Peptides binding to plastics can cause significant loss at low peptide concentrations	Minimize contact with plastics, excessive drying and storage at low concentrations (i.e., < 300fmol)
	Detection limits of the specific application	Ensure sample is within the detection limit of the specific downstream application – limits vary considerably based on application and instrumentation

## Related Thermo Scientific Products

<b>20062</b>	<b>Acetonitrile, 50mL</b>
<b>51101</b>	<b>Acetonitrile, 1L</b>
<b>53102</b>	<b>Trifluoroacetic Acid, HPLC grade, 10 × 1mL</b>
<b>89853</b>	<b>Phosphopeptide Isolation Kit</b>
<b>89866</b>	<b>2D Sample Preparation for Insoluble Proteins</b>
<b>89871</b>	<b>In-Gel Tryptic Digestion Kit</b>
<b>89895</b>	<b>In-Solution Tryptic Digestion and Guanidination Kit</b>
<b>89870</b>	<b>PepClean C18 Spin Columns, 25 columns each containing 8mg of C18 resin</b>
<b>89873</b>	<b>PepClean C18 Spin Columns, 50 columns each containing 8mg of C18 resin</b>

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<b>90035</b>	<b>CHCA, SA and DHB MALDI Matrix Sample Pack, Single-Use</b>
<b>90031</b>	<b>CHCA MALDI Matrix, Single-Use, 24 × 1mg</b>
<b>90032</b>	<b>SA MALDI Matrix, Single-Use, 24 × 1mg</b>
<b>90033</b>	<b>DHB MALDI Matrix, Single-Use, 24 × 4mg</b>

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