

## Making Connections

This note provides instructions on connecting standard-length 5 cm distal or standard coated PicoTips® to a transfer line or capillary column. “PicoTip” refers to any of New Objective’s high-quality tips for electrospray ionization, such as SilicaTips™, PicoFrits™, and TaperTips™.

NOTE: Users must take care when tightening the MicroTight® fittings, making sure to only tighten enough to prevent leaks from occurring. Due to the delicate nature of some fused-silica tubing, it is possible to damage the tubing if the fittings are overtightened.

### Loading the MicroTight® Union

- 1) Remove the MicroTight® Union from the PicoView® components box. Unscrew and remove the compression fittings from both ends of the union.
- 2) Screw the white gauge plug finger-tight on to one end of the union. Thread the fused-silica transfer line through a green MicroTight sleeve. The appropriate sleeve size is 0.002-0.003 inches greater than the OD of the capillary tubing. Use the green MicroTight sleeves with 360 um OD tubing. After the transfer line passes through a green sleeve, thread it through one of the compression fittings. Figure 2 shows the union loaded with the white gauge plug on the right and fused-silica tubing threaded through a tubing sleeve and a compression fitting. Cleave the end of the tubing and slip it into the union until both the tubing and the sleeve ends seat against the gauge plug inside the union. Screw the compression fitting finger-tight into the union, as shown in Figure 3.
- 3) Remove the gauge plug and return it to the PicoView components box.
- 4) Carefully trim a new green MicroTight sleeve to a length of approximately 14 mm, as shown in Figure 4. The shorter sleeve will allow the coating on the PicoTip to contact the conductive elastomer inside the CTM.
- 5) Choose a PicoTip from the assortment sent with PicoView. Although either coating style, the standard coating (-CE-) or the distal coating (-D-), will work, if flow rates permit, the distal coating is recommended due to its immunity to arcing.
- 6) Insert the back, or distal, end of the PicoTip through the trimmed sleeve and through the other compression fitting. When properly installed, the tip end should extend 15-20 mm past the end of the fitting when it is tight. This will afford optimal positioning of the PicoTip within the adjustment range of the stage plate. Using a ruler, measure and note the distance the tip extends from the fitting. Remove the fitting/sleeve/PicoTip assembly and carefully trim the back end of the PicoTip so the extension of the tip beyond the fitting is 15-20 mm. Cleave the remaining portion from the back end of the PicoTip. See Figures 5A and B.



Figure 1 MicroTight® union, gauge plug, and compression fittings

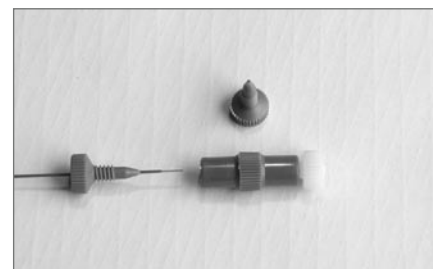


Figure 2 MicroTight® union with gauge plug and compression assembly ready to load



Figure 3 MicroTight® union with gauge plug and compression assembly loaded

- 7) After trimming, reinsert the assembly into the union, seat the PicoTip and sleeve against the transfer line or column tubing, and tighten the compression fitting finger-tight. Pull gently on the tubing to ensure the connection is tight. Figure 6 depicts the fully assembled union.

CAUTION: Always use chemical- and puncture-resistant gloves and ASNI-approved safety glasses when handling fused-silica tubing.

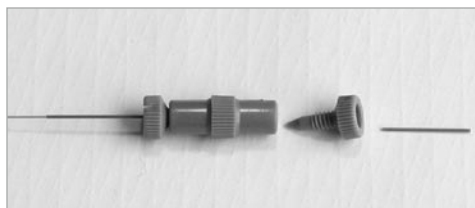


Figure 4 Union, compression fitting, and MicroTight® sleeve cut to 14 mm

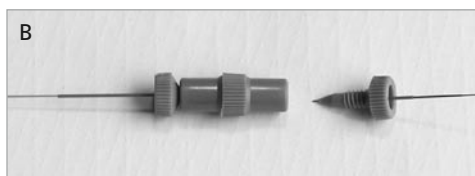
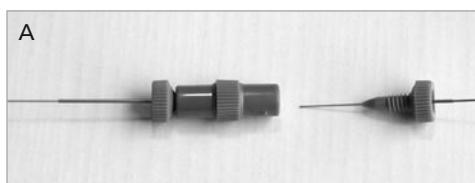


Figure 5 PicoTip™ in fitting, before (A) and after (B) trimming to length



Figure 6 A fully assembled MicroTight® union

### Using a PicoFrit® Column

PicoView® is ideal for performing nanoscale LC-MS and LC-MS/MS using PicoFrit® columns. In this configuration, the high voltage is applied to the back of the column through a liquid junction in the uncoated tip module. The PicoFrit column is packaged as a loop, as shown in Figure 7, to keep both ends of the column at the same pressure during storage and shipping. Do not remove either the orange-colored or the clear sleeve of snug-fitting PEEK™ and FEP (Teflon®) tubing holding the ends of the loop together until the column is being prepared for installation and use.

There are some important considerations in the handling and use of PicoFrit columns that make them very different from any other chromatography columns on the market today. PicoFrit columns are fabricated from 360 um OD, polyimide-coated, fused-silica tubing. The column has a specially tapered tip with an integral high-porosity frit. Behind the frit is the packed chromatography bed. There is no frit at the back end of the bed, only unpacked fused-silica tubing. Mobile phase flow must always be directed toward the tip. Reversing the flow may result in partial or complete unpacking of the chromatography bed.

The column is shipped filled with methanol and needs to be conditioned with an appropriate mobile phase for your gradient. Since there is no distal frit, the distal end of the column bed may loosen with time; this is not a problem, as the bed will repack when the column is pressurized.

Note the label attached to the opaque orange PEEK sleeve that slides along the length of the column. The arrow on the label points toward the tip end. The tip end may also be identified by the charred section of polyimide coating just prior to the tapered region of the tip.

#### Removing a PicoFrit® Column from the FEP Sleeve

Care must be taken to properly remove the column from the FEP (Teflon) sleeve to prevent damaging the fragile tip and frit. Use the free-sliding orange PEEK sleeve to push the clear FEP sleeve off the fused-silica tubing without damaging the tip.

- 1) Remove the distal end of the column from the FEP sleeve by either pulling it free of the sleeve or (preferably) by cleaving the fused-silica tubing near the terminus.
- 2) Slide the PEEK sleeve toward the tip end until it butts up against the FEP sleeve, as in Figure 8.
- 3) Orient the FEP sleeve vertically, with the tip facing toward the floor. This way, when the FEP sleeve slides free, it will fall toward the ground without damaging the tip.
- 4) Carefully push the PEEK sleeve against the FEP sleeve until the FEP sleeve falls off, as shown in Figure 9. This may take a LOT of force. You will have to move the PEEK sleeve about 3-5 mm. Grasp the column in one hand while pushing the sleeve off with the other hand.
- 5) Do not let the PEEK sleeve slide off the tip end. Instead, remove it by sliding it over the back end.

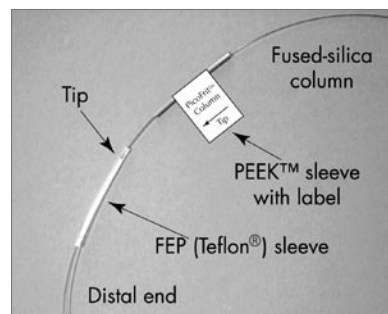


Figure 7 PicoFrit® column packaged for shipping

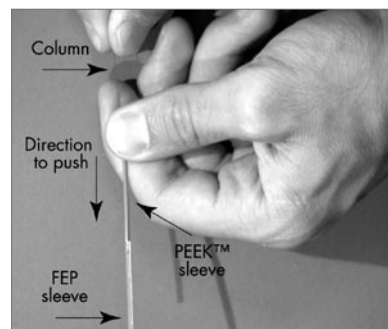


Figure 8 Pushing the PEEK™ sleeve against the FEP sleeve

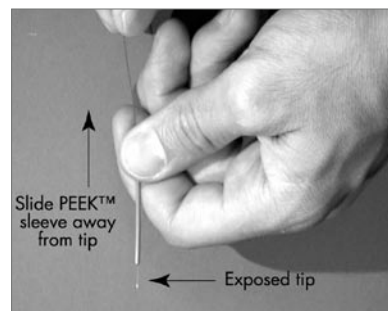


Figure 9 Pushing the FEP sleeve off the tip end of a PicoFrit® column

### Loading the MicroTight® Union

- 1) A MicroTight union within the tip holder is used as a pass through to position the tip toward the inlet.
- 2) Unscrew and remove the compression fittings from the end of the union. Thread the distal end of the PicoFrit® through a green MicroTight sleeve and through a MicroTight fitting. Gently screw the fitting into the union, but do not tighten. Push the distal end of the PicoFrit all the way through the union. The PicoFrit should freely move within the union.
- 3) When properly installed into the MicroTight union, the tip end should extend 15-20 mm past the end of the fitting when it is tight. This will afford optimal positioning of the tip within the adjustment range of the stage plate. Using a ruler, measure and note the distance the tip extends from the fitting. Gently move the PicoFrit until the tip extends 15-20 mm past the end of the fitting. Finger-tighten the compression fitting to hold the tip in place. Figure 10 shows a correctly loaded union.

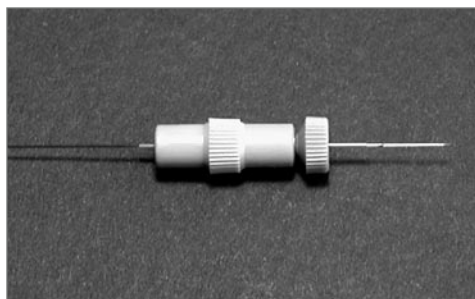


Figure 10 PicoFrit® column loaded into MicroTight union

### Supplying High Voltage

#### Plumbing the MicroTee

- 1) The MicroTee joins the transfer line to the PicoTip® and supplies the high voltage. Orient the MicroTee as shown in Figure 11 so that the platinum electrode is facing away from the user and the setscrews are visible. Unscrew the nuts and remove the black MicroFerrules from the posts of the MicroTee.

**WARNING:** Do not loosen the setscrews or remove the electrode cap, as this may damage the electrode. The solvent will not become charged and an electrospray will not form.

- 2) Thread the end of the PicoTip tubing through a green MicroTight sleeve, which is used for assembly with 360 um OD tubing. Make sure the PicoTip does not extend past the tubing sleeve end that will be inserted into the MicroTee. Thread the sleeved PicoTip through the fitting nut and a black MicroFerrule, as shown in Figure 12.
- 3) Cleave the end of the PicoTip after the tubing is threaded through the sleeve, nut, and ferrule. (Refer to Tech Note FS-1 for cleaving technique.) Slip the end of the tubing through the right post of the MicroTee, as viewed in Figure 13A, until the tubing and sleeve seat against the bottom ledge inside the post, as shown in Figure 13B. Screw the nut finger-tight onto the MicroTee.



Figure 11 MicroTee with sleeves

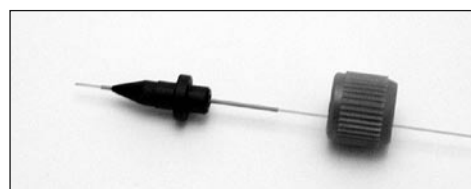


Figure 12 Fitting nut, MicroFerrule, and sleeve threaded by PicoTip

- 4) Insert the distal end of the fused-silica transfer line through a green MicroTight sleeve, then through the nut and the black MicroFerrule, as shown in Figure 14A. Carefully trim the end of the transfer line. After trimming, insert the assembly back into the MicroTee, seat the transfer line, ferrule, and sleeve against the PicoTip, and finger-tighten the nut, as shown in Figure 14B. Gently pull on the tubing ends to ensure the connection is tight. Check for leaks by running solvent through the tubing at the expected operating pressure. Leaks will be apparent if solvent collects at the exposed ends of the sleeves.

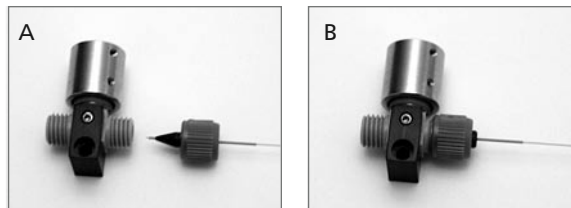


Figure 13 Assembling the nut, ferrule, sleeve, and PicoTip® (A) and securing finger-tight into the MicroTee (B)

### Plumbing the ZDV Union

NOTE: This option is not recommended with PicoFrit® columns.

- 1) Remove the ZDV union from the PicoView components box. Orient the union as shown in Figure 15 so that the “T” bracket is facing away from the user. Unscrew the nuts and remove the ferrules from the union.
- 2) Insert a green SealTight™ sleeve through the ferrule and nut. Thread a PicoTip through the sleeve/ferrule/nut assembly, as shown in Figure 16A. Cleave the end of the PicoTip after the tubing is threaded through the sleeve, ferrule, and nut. Slip the end of the tubing into the right side of the union, as viewed in Figure 16B, until the ferrule and sleeve seat against the bottom ledge inside the union. Screw the nut finger-tight into the union. Do not tighten enough to compress the sleeve.



Figure 15 ZDV union assembly

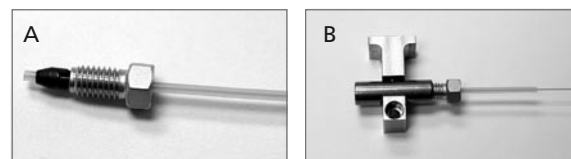


Figure 16 Assemble the nut, ferrule, sleeve, and PicoTip (A) and secure finger-tight into union (B)

NOTE: Be careful not to touch the tip to any surface.

- 3) Trim 5 mm from the end of a green SealTight™ sleeve and insert the sleeve through the ferrule and nut. Thread the back end of the fused-silica transfer line through the sleeve/ferrule/nut assembly. Cleave the back end of the transfer line and slip it into the left side of the union until the transfer line/ferrule/sleeve assembly seats against the PicoTip. Tighten the nuts on both ends of the union. Pull gently on the tubing to ensure the connections are tight. Figure 17 depicts a fully assembled ZDV union.

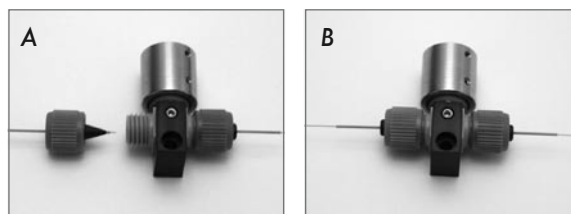


Figure 14 (A) Assembling the nut, ferrule, sleeve, and transfer line and (B) securing into the MicroTee

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