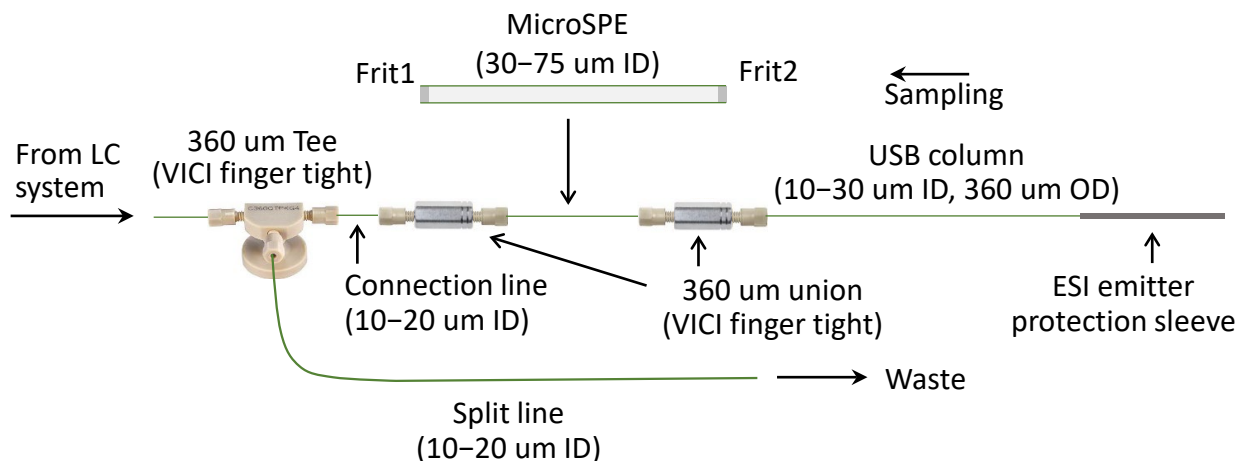


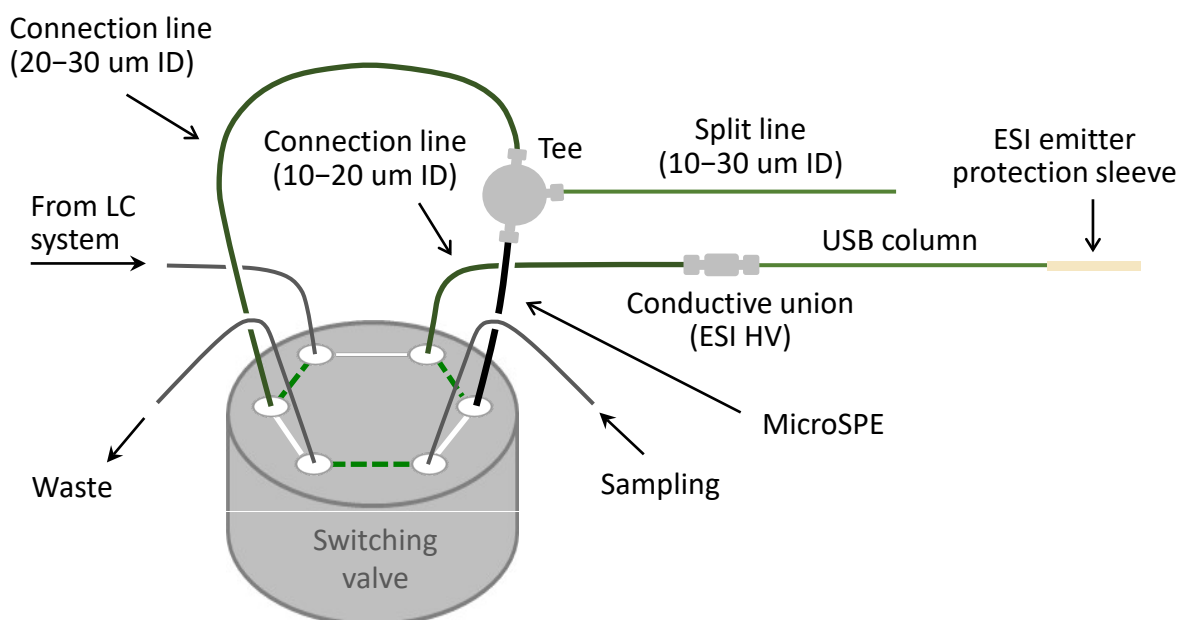
# Installation of CoAnn 10-30 $\mu\text{m}$ ID USB NanoLC Columns in a NanoLC-MS System

Manual operation:



Operation: Load a sample on the microSPE and connect the loaded end of the microSPE to the USB column inlet via a 360  $\mu\text{m}$  union (VICI), connect the other end of the microSPE to another union where ESI HV is applied, and connect the union to a 360  $\mu\text{m}$  tee (VICI) via a connection line (360  $\mu\text{m}$  OD fused silica capillary tube). Connect a split line to the tee and control the split flow with the length of the split narrow fused silica capillary tube. The flow slit can be removed if LC pumps accurately output mobile phase flows small enough for operating the USB column.

Automatic operation:



Note: If a nanoLC system can accurately output mobile phase flows small enough for operating a USB column the flow split can be removed by using a union to connect the microSPE and the connection line.

The setup can also be used for routine 50–75  $\mu\text{m}$  ID nanoLC–MS analysis (typically without need of flow split).

#### References:

1. Y Shen, R Zhao, SJ Berger, GA Anderson, N Rodriguez, RD Smith. High-efficiency nanoscale liquid chromatography coupled on-line with mass spectrometry using nanoelectrospray ionization for proteomics. Analytical chemistry 74 (16), 4235-4249.
2. Y Shen, N Tolić, C Masselon, L Paša-Tolić, DG Camp, KK Hixson, R Zhao, ...Ultrasensitive proteomics using high-efficiency on-line micro-SPE-nanoLC-nanoESI MS and MS/MS. Analytical chemistry 76 (1), 144-154.
3. Y Zhu, PD Piehowski, R Zhao, J Chen, Y Shen, RJ Moore, AK Shukla, ... Nanodroplet processing platform for deep and quantitative proteome profiling of 10–100 mammalian cells. Nature communications 9 (1), 1-10.