UltraMicroSpin™ (2-25µL elution volume, 3-30 µg max. capacity)* and *MicroSpin™ Columns (5-50µL elution volume, 6-60 µg max. capacity)

*Directions for Reversed Phase **(RPC):(p/n: SUM SS04V- - SS18V & SEM SS04V- - SS18V)**:*

These spin columns of C4 or C18 will retain non-polar solutes such as peptides, proteins, and detergents. Salts, and polar solutes like DNA will not be retained. This permits the removal of SDS from samples prior to mass spectrometry, removal of toxic substances prior to bioassay, and preliminary fractionation of a mixture by hydrophobic differences. Use of 1.0% TFA will increase the binding of peptides and proteins.

- Slide the adapter collar onto the spin column and place it in a 2ml micro centrifuge tube.
- **Conditioning the column**: Pipette 100µl of conditioning solvent (e.g., 100% acetonitrile or MeOH) into the column and centrifuge it for 1 min. at about 110x g (@ ~800 rpm with an Eppendorf micro centrifuge). Flush with 2 bed volumes (50 or 100µl, respectively) of 100% water. Remove the collecting tube and blot dry any moisture on the exterior of the column.
- **Processing the sample**: Add your 2-100 µl of sample to the column and place it in a new 2ml centrifuge tube. Spin the tube 1 min. at 110 x g. Peptides, proteins and detergents will be retained, while, salts, and polar solutes, like DNA, will elute into the liquid in the collecting tube. Discard this liquid unless these are the molecules you are after. Add an additional 25 or 50µl of equilibration buffer and repeat the spin to wash out any traces of salts from your sample. Repeat once again if necessary.
- **Releasing the sample**: Add 2-50µl of 80% MeCN or MeOH to the tube, preferably containing 25mM formic acid or some other volatile electrolyte. Spin as above. Peptides and proteins will be in the liquid in the collection tube. If a sample is especially non-polar (See Note A below), it may be necessary to repeat this step to elute all of the sample. If especially polar, then bind and equilibrate in 100% water containing 0.1% TFA or use the more retentive (for hydrophilic cpds.), water wettable column, TARGA C18 (p/n SUM SS18R).

NOTES:
- Columns can be reused by washing three times with two bed volumes (50µL, 100µL or 500µL, respectively) of 100% MeCN, MeOH or *n*-PrOH containing 25 mM formic acid (aq.) and then washing three times with two bed volumes of loading or equilibration buffer.
- **Sample composition**: *Important*: The sample and the equilibration buffer should contain comparable amounts of acetonitrile (e.g., 0 - 5%). Otherwise, polar solutes such as peptides and proteins might not be retained. Including 1.0% TFA increases binding capacity for peptide capture. Decrease the organic solvent concentration of the sample if yields are low.

MacroSpin™ Columns (50-150µL elution volume, 30-300 µg capacity)

*Directions for Reversed Phase **(RPC): (p/n: SMM SS04V - -SS18V)**:*

- **Conditioning the column**: Pipette 400µl of conditioning solvent (e.g., 100% acetonitrile or MeOH) into the column and centrifuge it for 1 min. at about 110 x g (@ ~800 rpm with an Eppendorf micro centrifuge). Flush with 2 bed volumes (400µl) of 100% water. Remove the collecting tube and blot dry any moisture on the exterior of the column.
- **Processing the sample**: Add your 50-450µl of sample to the column and place it in a new 2ml centrifuge tube. Spin the tube 1 min. at 110 x g. Peptides, proteins and detergents will be retained, while, salts, and polar solutes, like DNA, will elute in the liquid in the collecting tube. Discard this liquid unless these are the molecules you are after. Add an additional 50-150µl of loading or equilibration buffer and repeat the spin to wash out any traces of salts from sample of interest. Repeat once again if necessary.
- **Releasing the sample**: Add 50 - 150µL of 80% MeCN or MeOH to the tube, preferably containing 25mM formic acid or some other volatile electrolyte. Spin as above. Peptides and proteins will be in the liquid in the collection tube. If a sample is especially non-polar (See Note A above), it may be necessary to repeat this step to elute all of the sample. If especially polar, then bind and equilibrate in 100% water containing 0.1% TFA or use the more retentive (for hydrophilic cpds.), water wettable column, TARGA C18 (p/n SMM SS18R).

96-Well Spin and 96-Well MACROSpin RPC Plates (10-60µL elution volume, 10-100 µg max. capacity and 40-120µL elution volume, 40-400 µg max. capacity, respectively).

*Directions for Reversed Phase: **(p/n: SNS SS04V- - SS18V, SNS or SS04V-L - - SS18V-L)***

- Tap the column gently to ensure that the dry column material is settled at the bottom of the columns and condition as above. Foil is for sealing purposes only. All 96 wells do not need to be opened at the same time. Remove foil from as many rows as desired for your application. Foil should be cut with a razor or other sharp blade.
- Place the 96-Well Spin Column into a collection plate and pipette 200µL of organic solvent (400µL for the MACROSpin Plates) into all opened wells and centrifuge the plate for 1 minute in the collection plate at 110x g to wet the RPC phase then repeat with 95-100% water to equilibrate.
- You can reuse the emptied collection plate for sample loading. Blot dry any liquid on the exterior of the column. Add your 50-100µL sample (50-150µL for the MACROSpin Plates) to the top of a well. Be careful to ensure that the sample is placed in the center of the well. Having 0.1% TFA in the sample can facilitate binding, but it isn't necessary.
- Place the column in a new collection plate when the appropriate elution solvent is added (i.e. higher concentrations of MeCN or organic solvent). Spin the plate for 1 minute at 110x g. After centrifugation, the purified sample will be in the collecting tube and will be ready for further use. If a sample is especially non-polar (See Note A above), it may be necessary to repeat this step to elute all of the sample. If especially polar, then bind and equilibrate in 100% water containing 0.1% TFA or use the more retentive (for hydrophilic cpds.), water wettable column, TARGA C18 (p/n SNS SS18R).

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