

APPLICATION NOTE

Trap Concentrate and Map, Using Acquity UPLC I class Plus

Identifying small amounts of chemicals in large volumes has been a time consuming and resource wasting process that is cluttering the relevant industries for some time.

Our aim is to make it automated, practical, relevant, reproducible and above all economical both in material as well as manpower to be on par with technological advances.

To this end two short narrow bore columns of 2.1 mm ID and 5 cm length are used with two 6 ports switching valves.

The first setup consists of having only the polymeric reversed-phase column online (SYROS® 1R Simulated-Monolith™) to trap 5 compounds in an aqueous solution at a concentration of 1 µg/ml.

The second setup would include a silica C18 column (Acquity UPLC® BEH C18 1.7 µm 2.1x50 cm column). To co-map the trapped compounds in the first polymeric column.

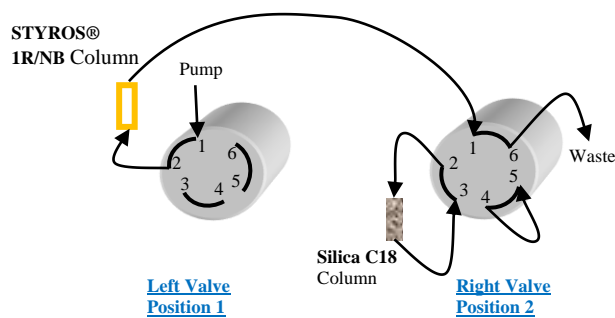
The buffers used consists of:

Solution A: DI H2O solution with the dissolved compounds.

Buffer B: 0.075 % TFA in DI H2O (for mapping)

Buffer C: 0.075 % TFA in ACN: H2O, 95: 5 (for mapping)

The following shows the starting position of the valves used:



Setup 1

In this first position, the Narrow Bore polymeric STYROS® 1R/NB column is used to trap the compounds in solution while discharging the effluent to waste.

The second setup switches the right valve to position 1 and bring the silica column online for co-mapping.

This prevents the excessive exposure of any Silica column to large volumes of aqueous solution.

On a UPLC Acquity I class Plus an Inlet Prerun step is programmed to run solution A with 5 compounds at a concentration of 1 µg/ml each in DI H2O.

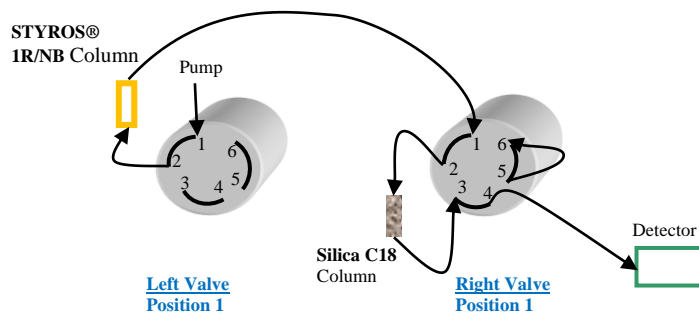
1-Trap the dissolved components on the Reversed phase polymeric STYROS® 1R/NB column as shown in Setup 1.

Time (Minutes)	% of Solution A	Flow rate (ml/min)
0		0
0.5	100	0.4

2-With both columns online as in Setup 2, a gradient is run to elute the trapped components.

Time	% of buffer B	% of buffer C	Flow rate ml/min
0			0
0.01	2	98	0.2
5	0	100	0.2
5.1	2	98	0.2
7	2	98	0.2

The 5 minutes gradient elutes all 5 components that were trapped on the first Narrow Bore column during the ½ minute initial run of the solution A.



Setup 2

An additional 2 minutes run is added to equilibrate back the column to its initial starting point for elution.

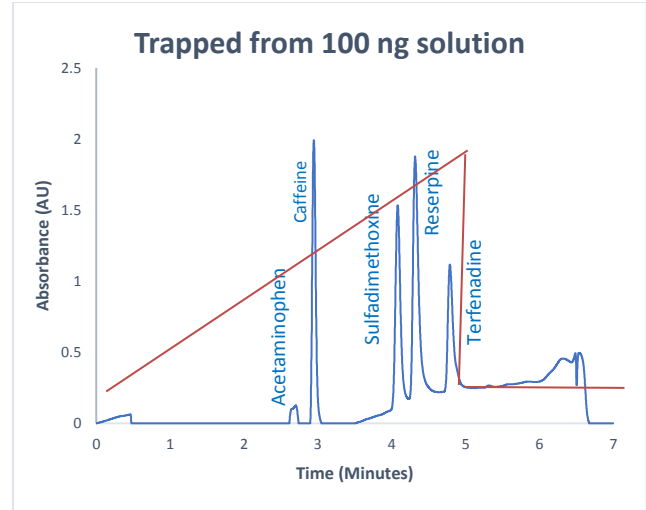
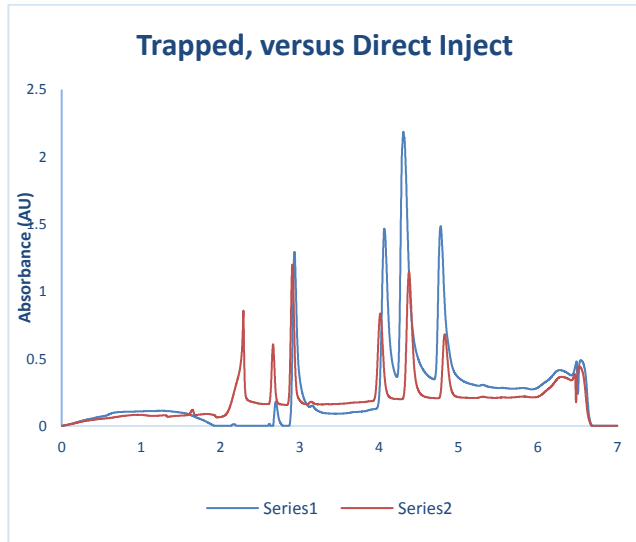
It is important to note the low consumption of solvents used in the process using Narrow Bore columns. (less than 2 ml)

The following overlaid chromatograms compares two injections:

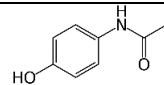
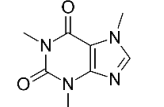
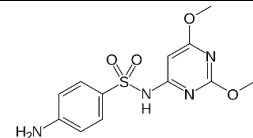
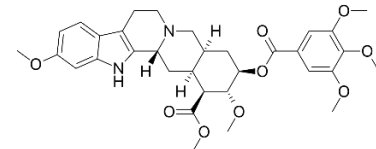
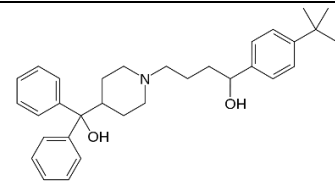
Series 1 is the run from the trapped components compared to Series 2 direct injection of the 5 components in the amount of 2 µl of a solution of 1 mg/ml of each in buffer B.

Further elution of the initial solution to 0.1 µg/ml, that is 100 ng/ml would require additional time to run the solution during the first step to trap enough material for mapping.

Time (Minutes)	% of Solution A	Flow rate (ml/min)
0		0
5	100	0.4



All 5 components have some solubility in water thus an interest in detecting them whereas compounds that do not dissolve in water would not be of interest as they would precipitate and not be washing away in water

	Acetaminophene M=151.16
	Caffeine M=194.19
	Sulfadimethoxine M=310.33
	Reserpine M=608.68
	Terfenadine M=471.67

