

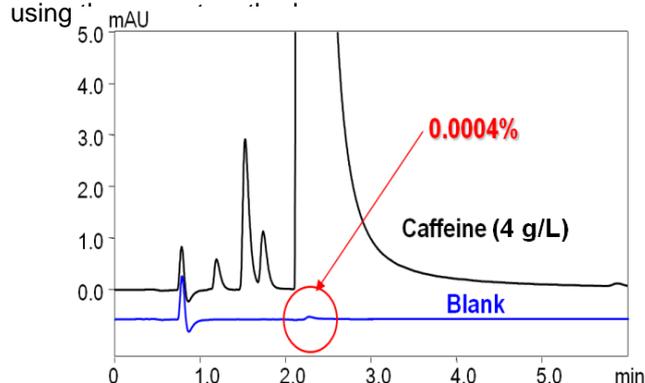
# Nexera Application Data Sheet No. 4

## The Lowest Near Zero Carryover

Nexera autosampler has been designed to provide our customers with the next generation tools for combating sample carryover on an entirely new level. It is now possible to perform a thorough rinse of the sample path and the needle surface by multiple different solvents eliminating the traces even of the stickiest of compounds. This is in addition to the flow-through needle design with minimized contact area which by itself significantly reduces carryover and insures highly reproducible analysis of trace compounds.

### Carryover of Caffeine

Without applying the injection port rinse, needle internal rinse and needle outer surface rinse caffeine injected at the concentration of 4 g/L produced a carryover of only 0.0004% in the subsequent injection of a blank (post-blank 1). The injection of a blank was followed by another blank injection (post-blank 2) at which point no carryover was detected using

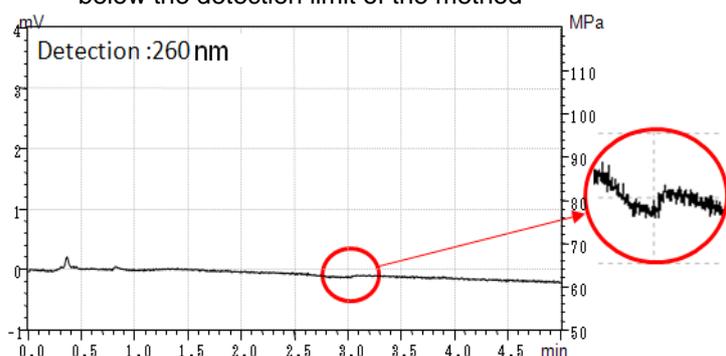


Injection	Carryover
Caffeine 4 g/L	-
Post-blank 1	0.0004%
Post-blank 2	Not Detected

Column : ODS (2.0 mm I.D. x 100 mm, 1.8  $\mu$ m)  
 Mobile Phase: : Methanol/Water = 2/8  
 Flow Rate : 0.4 mL/min  
 Column Temp. : 40  $^{\circ}$ C  
 Detection : UV 272 nm  
 Pressure : 100 MPa  
 Injection volume : 5  $\mu$ L

### Carryover of Chlorhexidine

Chlorhexidine is a well-known basic compound that is often used for carryover evaluation due to its highly adsorptive behavior with a wide range materials. In the following experiment 2 g/L chlorhexidine in water was introduced into the autosampler with the first sample injection. By the injection port rinse along with the needle inner and outer surface rinses it was possible to completely eliminate chlorhexidine carryover below the detection limit of the method



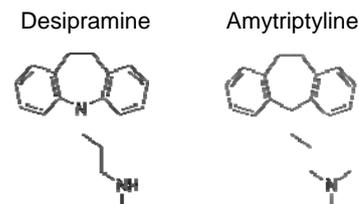
Injection	Carryover
Chlorhexidine 2	-
Post-blank 1	Not Detected ( $<0.003\%$ )

Column : ODS (2.0 mm I.D. x 100 mm, 1.8  $\mu$ m)  
 Mobile Phase: : Methanol/Water = 2/8  
 Flow Rate : 0.4 mL/min  
 Column Temp. : 30  $^{\circ}$ C  
 Detection : UV 272 nm  
 Pressure : 100 MPa  
 Injection volume : 5  $\mu$ L  
 Rinse solution : 0.05 % Formic acid in Methanol  
 Needle wash : Outer surface flush by rinse pump (1 s)  
                   : Needle dip rinse (0 s)  
                   : Needle internal rinse  
 Injection port rinse : Performed

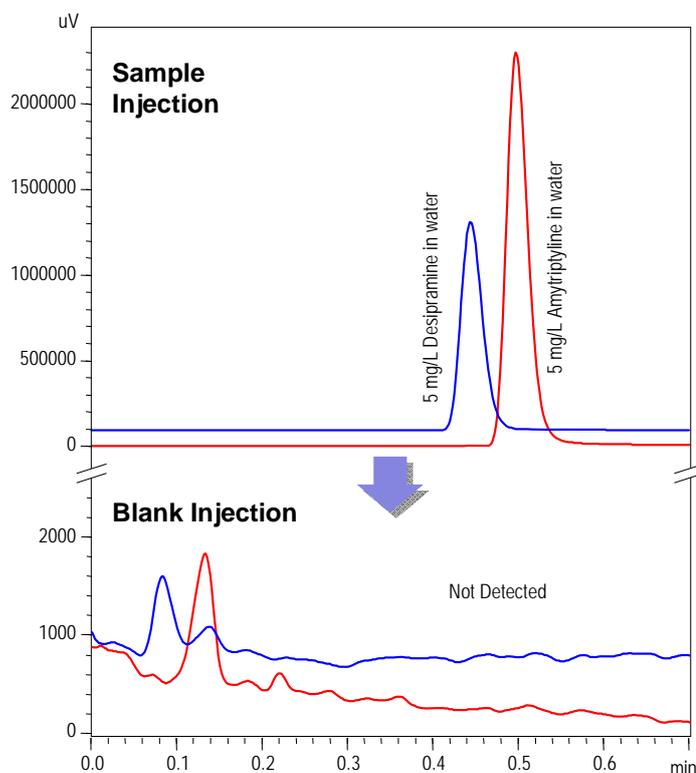
## LCMS-2020 Carryover

The single quadrupole mass spectrometer LCMS-2020 was used for an evaluation of carryover at the Nexera autosampler SIL-30AC. The chromatogram below shows an injection of a mixture of Desipramine and Amytriptyline at 5 mg/L each. The table below shows the comparable effectiveness of using different rinsing scenarios and the effect on carryover. By employing a needle dip rinse along with the needle pump rinse we were able to completely eliminate carryover of these compounds beyond the detection limit of the mass spectrometric method.

Nexera Rinse Setup	Desipramine Carryover	Amytriptyline Carryover
1. Needle Dip rinse 2. Needle Pump rinse 3. Needle Internal rinse 4. Injection Port rinse	Not Detected ( < 0.0042% or 1.0 pg )	Not Detected ( < 0.0027% or 0.68 pg )
1. Needle Dip rinse 2. Needle Pump rinse 3. Needle Internal rinse	Not Detected ( < 0.0042% or 1.0 pg )	Not Detected ( < 0.0027% or 0.68 pg )
1. Needle Dip rinse 2. Needle Pump rinse	Not Detected ( < 0.0042% or 1.0 pg )	Not Detected ( < 0.0027% or 0.68 pg )
1. Needle Dip rinse only	0.0098%	0.0078%
No rinse	0.052%	0.050%



Nexera/LCMS-2020



Column	: ODS (2.1 mm I.D. × 100 mm, 1.8 μm)
Mobile Phase	: A : 0.1% Formic acid B : Acetonitrile/Water (25/75)
Gradient	: B 25%→50% (0.75 min) →90% (0.76 min) →25% (3.01 min)
Flow Rate	: 1.5 mL/min
Column Temp.	: 40 °C
Injection Volume	: 5 μL
Detection	: LCMS 2020-ESI (+)
Pressure	: 95 MPa
Needle Dip Rinse	: 0.1% Formic acid in Methanol or Acetonitrile
Needle Pump Rinse	: 0.1% Formic acid in Acetonitrile
Needle Internal Rinse	: 0.1% Formic acid in Methanol
Injection Port Rinse	: 0.1% Formic acid in Methanol