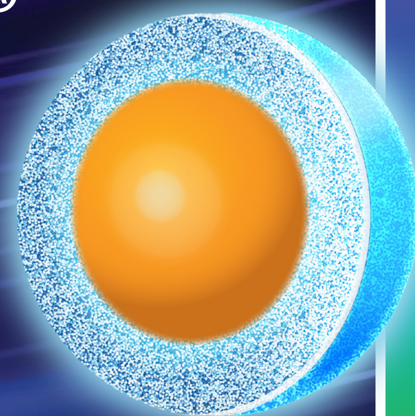
HALO®

ELEVATE

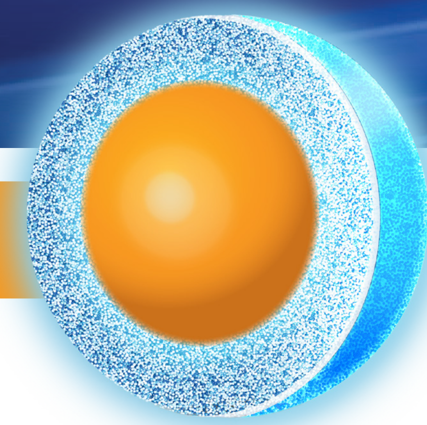


ENGINEERED FOR HIGH PH
PERFORMANCE ACROSS THE RANGE

ELEVATE

HALO® ELEVATE

QUALITY BY DESIGN



INTRODUCING HALO® ELEVATE

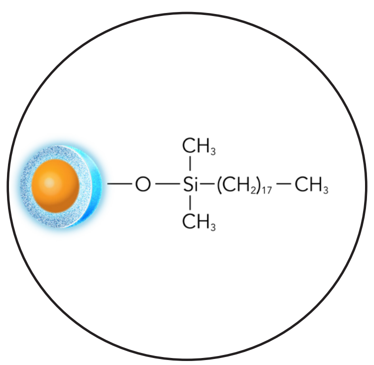
Built upon proven Fused-Core® particle technology for speed and efficiency, the HALO® ELEVATE product family incorporates surface modified organo-silane technology for alkaline resistance resulting in excellent stability in high pH environments.

With a wide operational use range of pH 2-12, HALO® Elevate allows for robust method development and improved separations for basic compounds that may present problems such as poor peak shapes, inadequate retention or limited load tolerance at low pH. Ideal for use with high pH mobile phases.

FEATURES OF HALO® ELEVATE

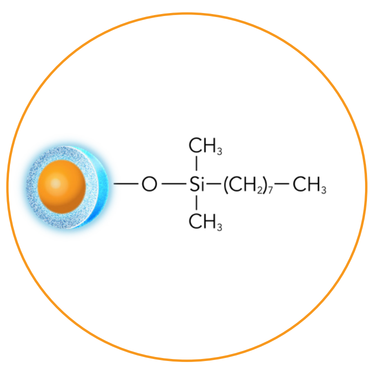
- THREE PHASE OFFERINGS:
  - C18, C8 and PHENYL-HEXYL Phases
- EXCELLENT STABILITY for high pH, high temperature environments
- THREE PARTICLE SIZES: 2, 2.7 and 5 µm
- ROBUST COLUMN OPTION To work the full range of operating conditions for separation selectivity of acids, bases, neutrals and zwitterions
- PROVEN HALO® FUSED-CORE® TECHNOLOGY for uniform column loading, quality separations, speed and ruggedness

PRODUCT CHARACTERISTICS



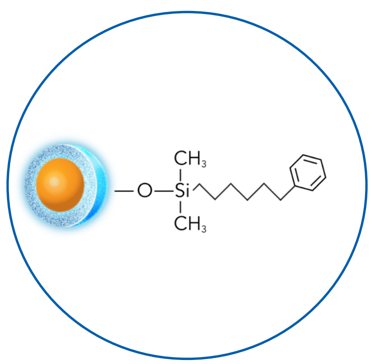
C18

Ligand: dimethyloctadecyl
Particle Size: 2, 2.7 and 5µm
Pore Size: 120 Å
USP Designation: L1
*Reported: 2, 2.7, 5µm respectively
Carbon Load*: 5.2%, 5.6%, 4.5%
Surface Area*: 70, 75, 60 m²/g
Endcapped: YES
pH Operating Range: 2-12
Temp Limit: 60 °C
Pressure Stability*: 1000, 600, 600



C8

Ligand: dimethyloctyl
Particle Size: 2, 2.7 and 5µm
Pore Size: 120 Å
USP Designation: L7
*Reported: 2, 2.7, 5µm respectively
Carbon Load*: 3.8%, 4.2%, 3.2%
Surface Area*: 70, 75, 60 m²/g
Endcapped: YES
pH Operating Range: 2-12
Temp Limit: 60 °C
Pressure Stability*: 1000, 600, 600



PHENYL-HEXYL

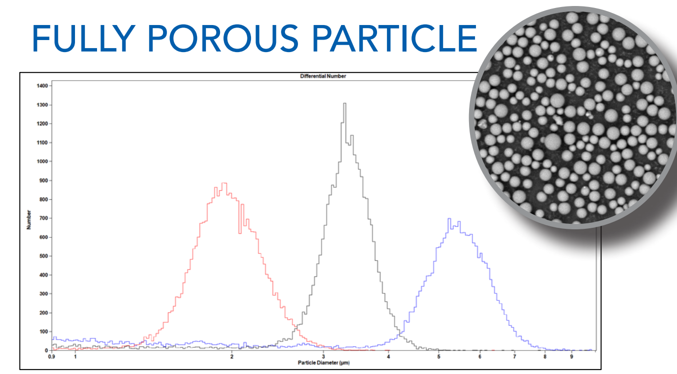
Ligand: dimethylphenylhexyl
Particle Size: 2, 2.7 and 5µm
Pore Size: 120 Å
USP Designation: L11
*Reported: 2, 2.7, 5µm respectively
Carbon Load: 4.5%, 5.1%, 4.0%
Surface Area*: 70, 75, 60 m²/g
Endcapped: YES
pH Operating Range: 2-12
Temp Limit: 60 °C
Pressure Stability*: 1000, 600, 600

FUSED-CORE® PARTICLE SIZE CONSISTENCY

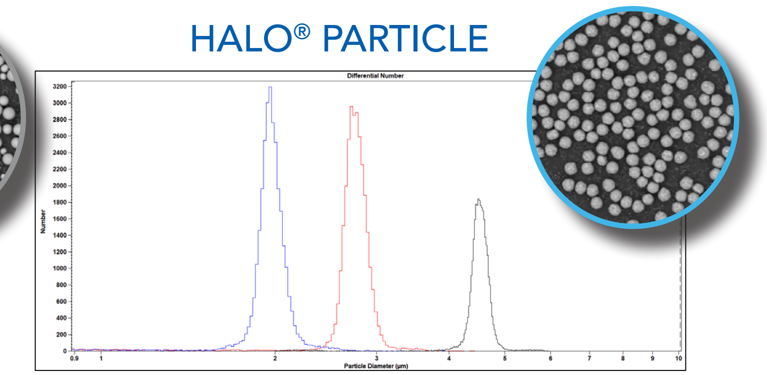
The quality of AMT products stems from its complete control over the entire manufacturing process – from the start of high quality raw materials to the final particle, quality controls and measurements are built into every process. Cores and shells are carefully controlled resulting in tight particle size distributions for optimized loading parameters that culminates to a highly efficient performance packed column.

The tight control of particle size distribution shown in the following figure represents an impressive 50% lower stdev of any product size compared to a traditional fully porous product currently on the market. This result contributes greatly to our commitment for customers' long-term repeatability of separations utilizing AMT powered HPLC columns.

FULLY POROUS PARTICLE

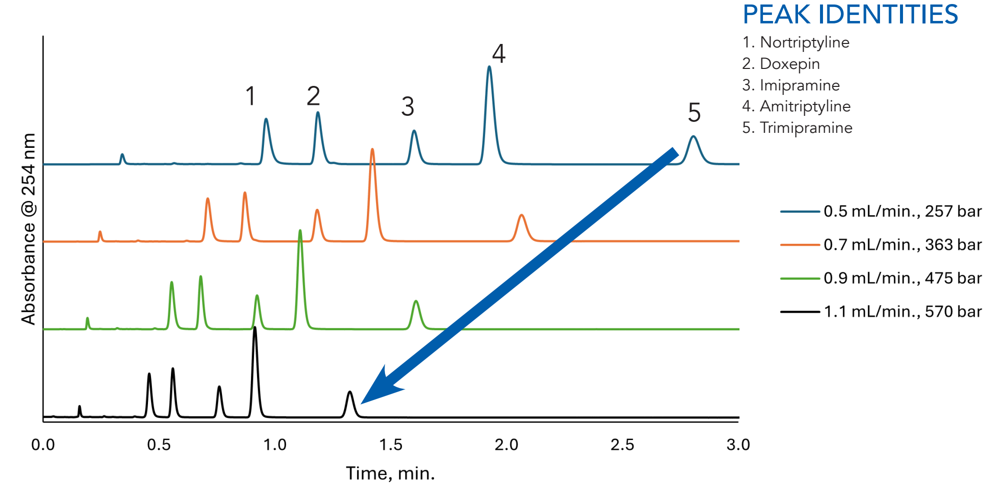


HALO® PARTICLE



ROBUST AT FASTER FLOW RATES

Fused-Core® (superficially porous) particles deliver UHPLC-like efficiency at lower backpressure, enabling higher flow rates and faster HPLC analyses without compromising separation quality. For example, a tricyclic antidepressant separation at pH 10 can be run at ***more than double the optimal flow rate***, maintaining high efficiency while staying below the 600 bar pressure limit.



PEAK IDENTITIES

1. Nortriptyline
2. Doxepin
3. Imipramine
4. Amitriptyline
5. Trimipramine

TEST CONDITIONS

Column: HALO 120 Å ELV C18, 2.7 µm, 2.1 x 100 mm
Mobile Phase A: 10mM Ammonium Bicarbonate, pH 10
Mobile Phase B: Acetonitrile
Isocratic: 60% B
Temperature: 35 °C
Sample Solvent: 50/50 Acetonitrile/ Water
Wavelength: PDA, 254 nm
Flow Cell: 1 µL
Data Rate: 100 Hz
Response Time: 0.025 sec.
LC System: Shimadzu Nexera X2

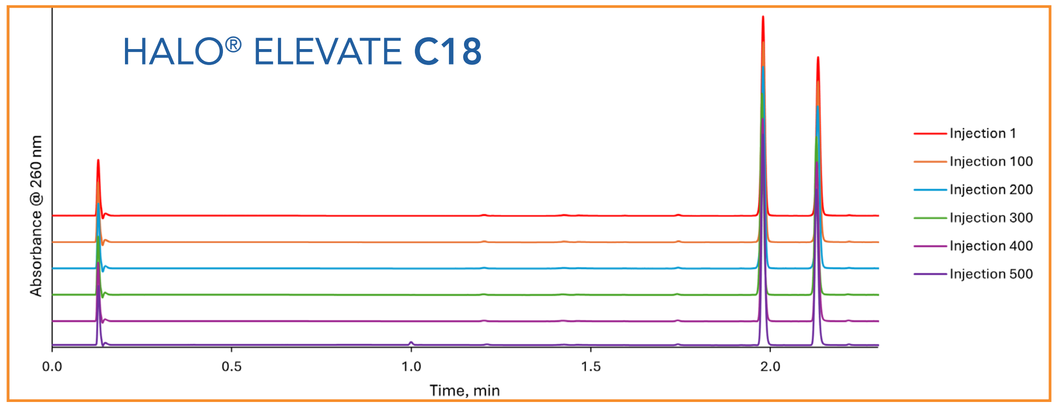
HIGH TEMPERATURE/ PH AMITRIPTYLINE STABILITY USING HALO® ELEVATE

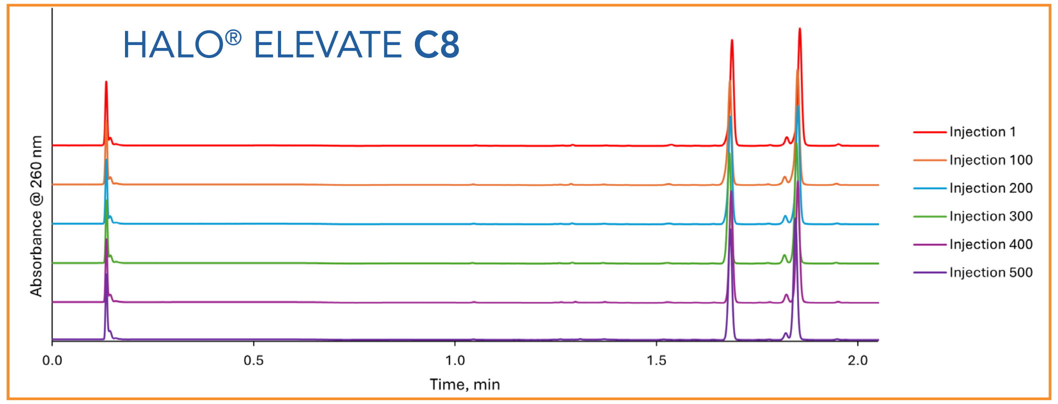
Unlike standard silica columns, HALO® ELEVATE columns operate reliably between pH 2–12 and at elevated temperatures. The HALO® ELEVATE column demonstrates this capability, maintaining stable retention (<1% change) and consistent backpressure at pH 10 and 60 °C over 20,000 column volumes.

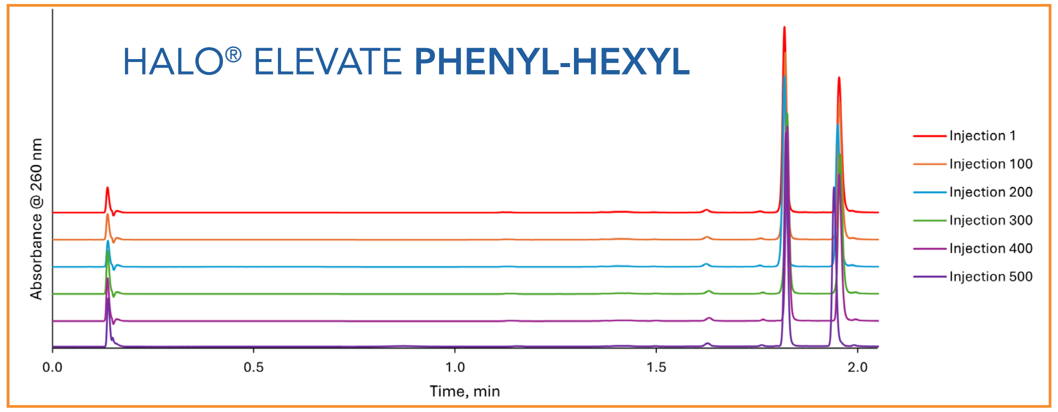
Column: 2.7 µm, 2.1 x 50 mm (phase as labeled)
Mobile Phase A: 95/5 10mM Ammonium Bicarbonate, pH:10/ ACN
Mobile Phase B: Acetonitrile
Flow Rate: 0.8 mL/min
Back Pressure: 220 bar
Temperature: 60 °C

Gradient: Time	%B
0.0	0
2.5	95
3.0	95
3.1	0
5.0	0

Detection: UV/PDA, 260 nm
Injection Volume: 1.0 µL
LC System: Shimadzu Nexera X2





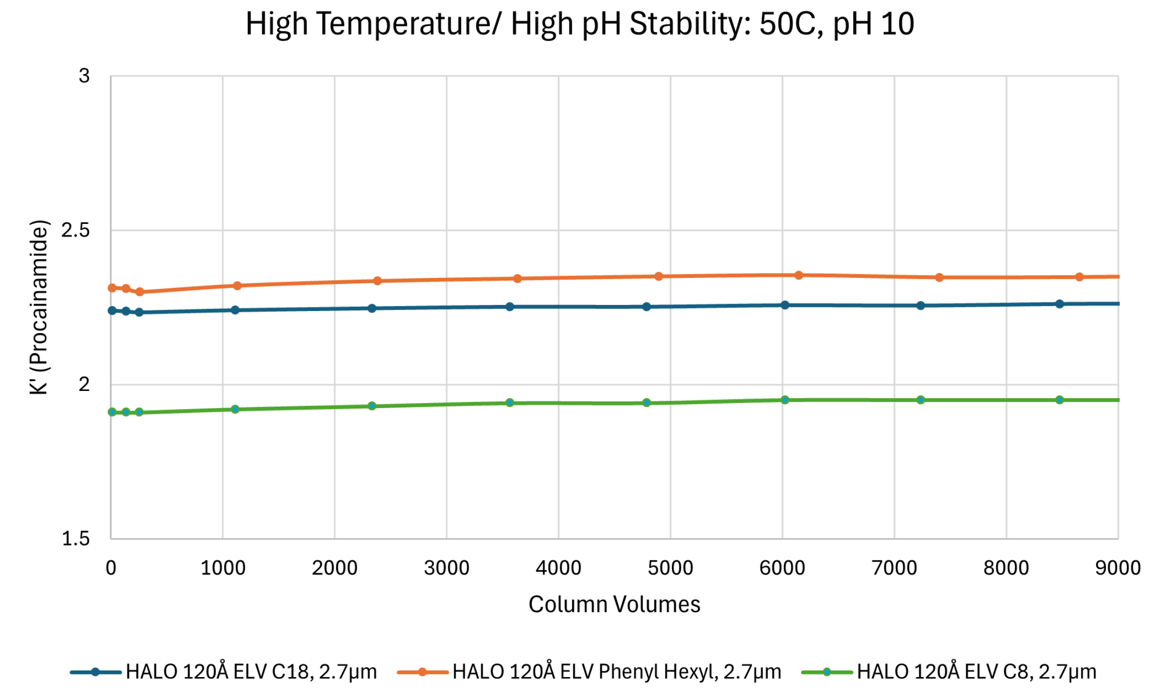


PEAK IDENTITIES

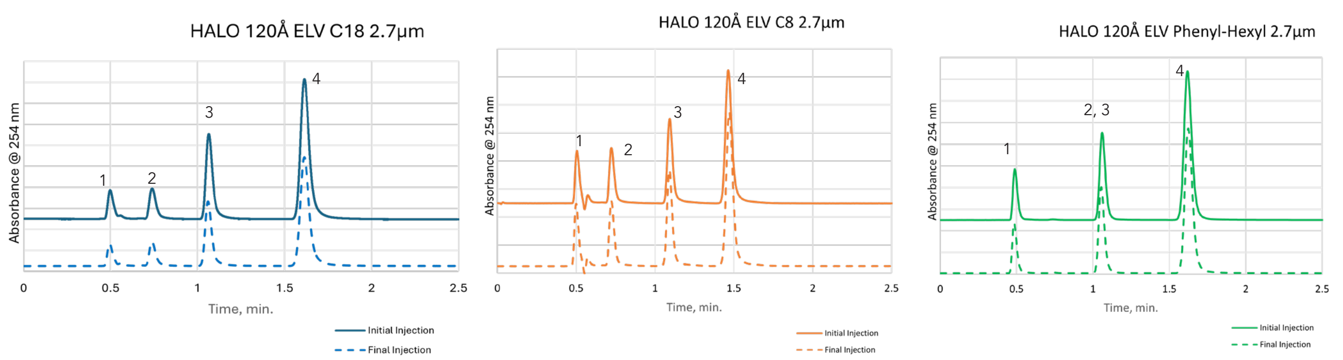
1. Uracil
2. Acenaphthene
3. Amitriptyline

HIGH PH STABILITY TESTING @ 50 C

A combination of neutrals and bases are observed on a HALO® ELEVATE C18, C8, and Phenyl-Hexyl column showing excellent reproducibility over 9,000 column volumes. Separation is performed under high temperature (50°C), and high pH (10), isocratic conditions.



Below are the chromatograms that were included in the stability run above. A chromatogram from beginning and end of the run can be found below for each product chemistry all demonstrating excellent stability of performance over time.



TEST CONDITIONS

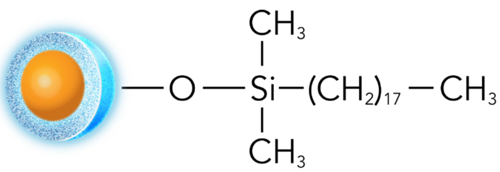
Column: HALO 120 Å ELV C18/C8/Phenyl-Hexyl, 2.7 µm, 2.1 x 50mm
Mobile Phase A: Ammonium Bicarbonate, pH 10/ Water
Mobile Phase B: Methanol
Isocratic: 30% B
Flow Rate: 0.21 mL/min
Temperature: 50 °C
Injection Volume: 0.3 µL
Sample Solvent: Water
Detection: UV, 254nm
Instrument: Shimadzu Nexera

PEAK IDENTITIES

1. Uracil
2. Caffeine
3. Acetanilide
4. Procainamide

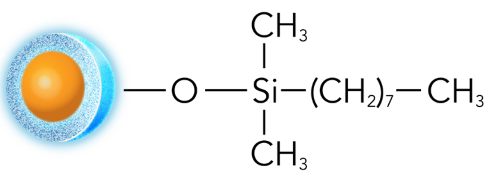
ELEVATE SELECTIVITY PORTFOLIO

HALO® ELEVATE C18



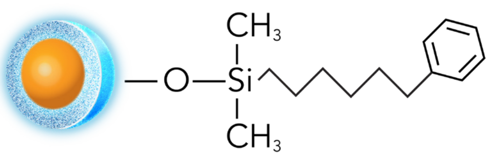
- Retention mainly governed by hydrophobicity, can offer other attributes but to alesser extent
- RPLC workhorse for majority of routine assays
- Universal phase for acids, bases and neutral solutes

HALO® ELEVATE C8



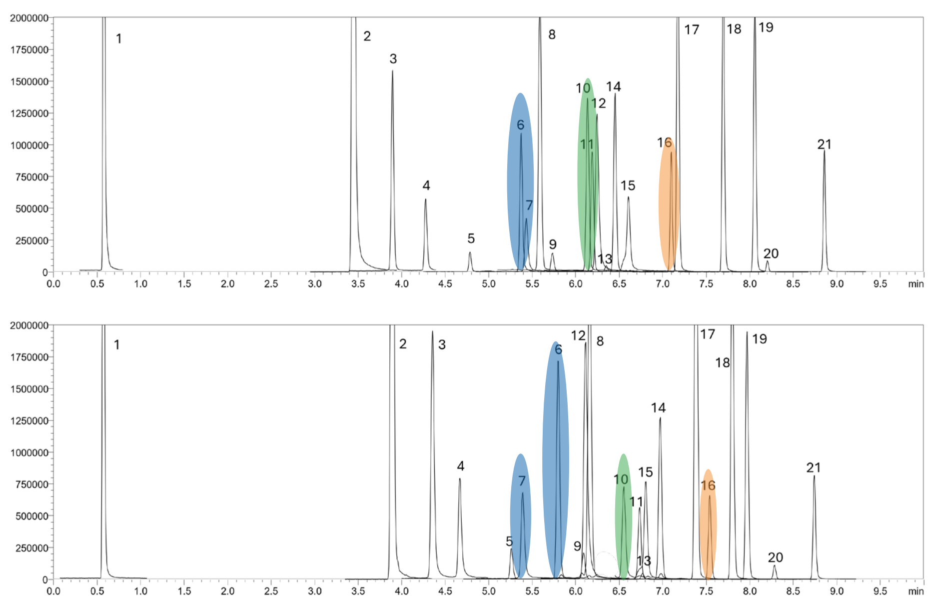
- Less hydrophobic compared to C18 and significant lower acidic ion exchange capabilities compared to C18
- Ideal for broad range of analytes

HALO® ELEVATE PHENYL-HEXYL



- Less hydrophobic than C18
- Ideal for separation of aromatic compounds. Phenyl moiety offers pi-pi interactions – with the highest H-bonding capacity
- Significantly decreased acidic ion exchange and slightly higher total ion exchange (pH>7) compared to C18

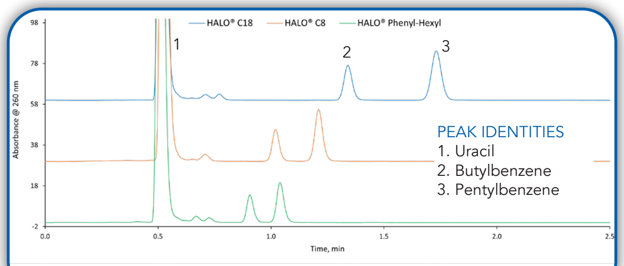
This panel of 21 drugs and metabolites run via LC-MS/MS demonstrates selectivity differences between ELEVATE C18 and ELEVATE Phenyl-Hexyl. Several peaks are highlighted to demonstrate the value of selectivity differences depending on the peak of interest. HALO® ELEVATE is offered in three phases to allow for peak selectivity differences.



PEAK IDENTITIES

1. Ecgonine
2. Benzoylecgonine
3. Oxymorphone
4. Noroxycodone
5. 6-Acetylmorphine
6. Levamisole
7. Amphetamine
8. Naloxone
9. Hydrocodone
10. Oxycodone
11. Alpha-Hydroxyalprazolam
12. Methamphetamine
13. Heroin
14. Alprazolam
15. Xylazine
16. Diazepam
17. Cocaine
18. Cocaethylene
19. Diphenhydramine
20. Fentanyl
21. Methadone

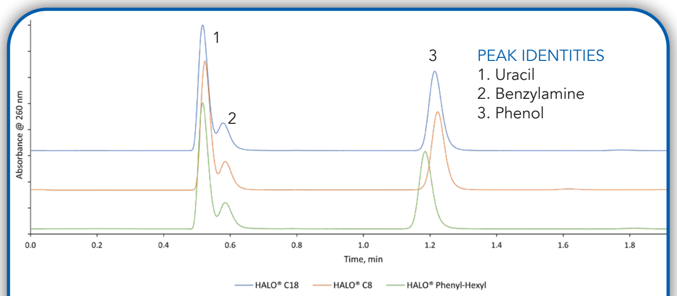
The HPLC Tanaka test is a standardized characterization protocol used to evaluate the selectivity and performance of reversed-phase (RP) columns. It uses a set of specific chemical probes to measure how a stationary phase interacts with different types of molecules. The results are typically visualized using a hexagonal radar plot (often called a "Tanaka Plot"), where each axis represents a different chromatographic property. Below shows a "Euerby-modified" test of the three HALO® phases among each other to illustrate some anticipated differences one can consider when selecting the right phase.



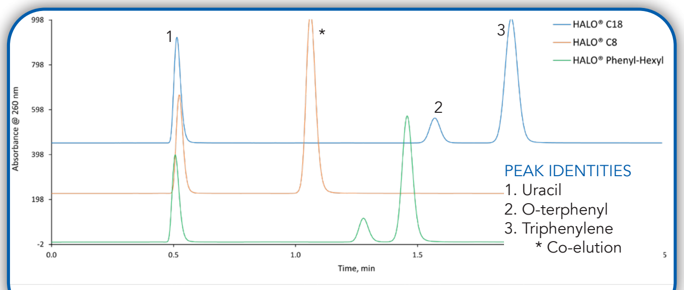
80/20 Methanol/ Water, 40°C

Hydrophobic Retention (k'):

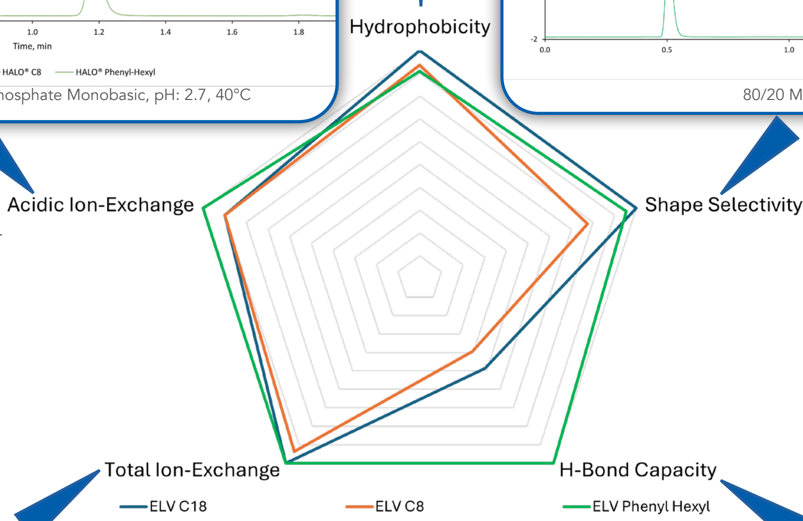
Measures the hydrophobicity and surface area available for retention.



30/70 Methanol/ 20mM Potassium Phosphate Monobasic, pH: 2.7, 40°C



80/20 Methanol/ Water, 40°C

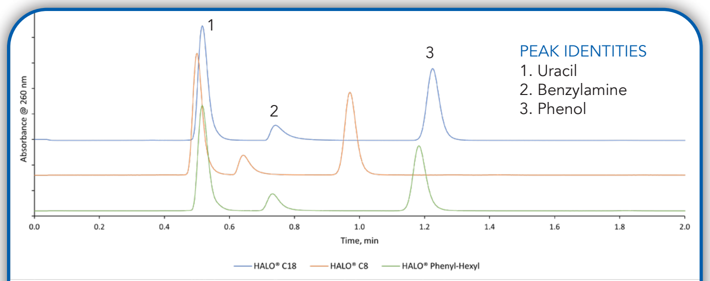


Ion Exchange Capacity (Low pH):

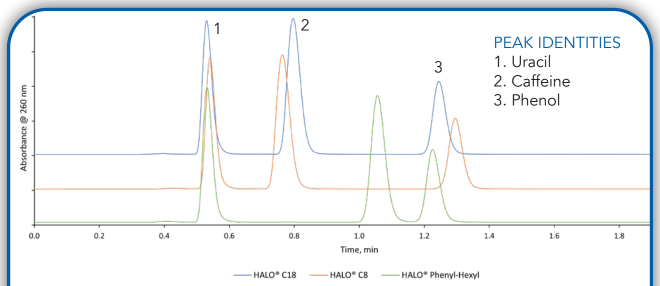
Measures acidic silanol activity or metal impurities at pH 2.7.

Steric (Shape) Selectivity:

Assesses the column's ability to separate molecules with the same elemental composition but different 3D shapes. Measured using the separation factor (α)



30/70 Methanol/ 20mM Potassium Phosphate Dibasic, pH: 7.6, 40°C



30/70 Methanol/ Water, 40°C

Ion Exchange Capacity (High pH):

Measures total free silanols through electrostatic attraction with basic molecules at pH 7.6.

Hydrogen Bonding Capacity:

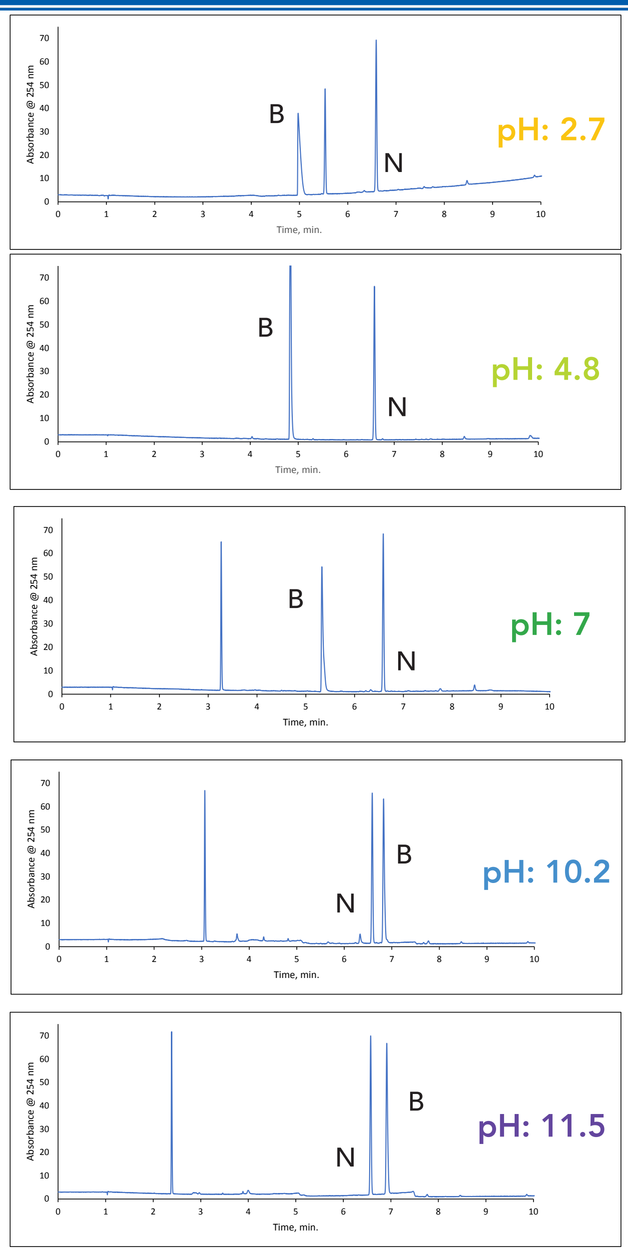
Indicates the presence of available silanol groups and the column's tendency to form hydrogen bonds.

ACHIEVING SELECTIVITY DIFFERENCES & LEVERAGING pH

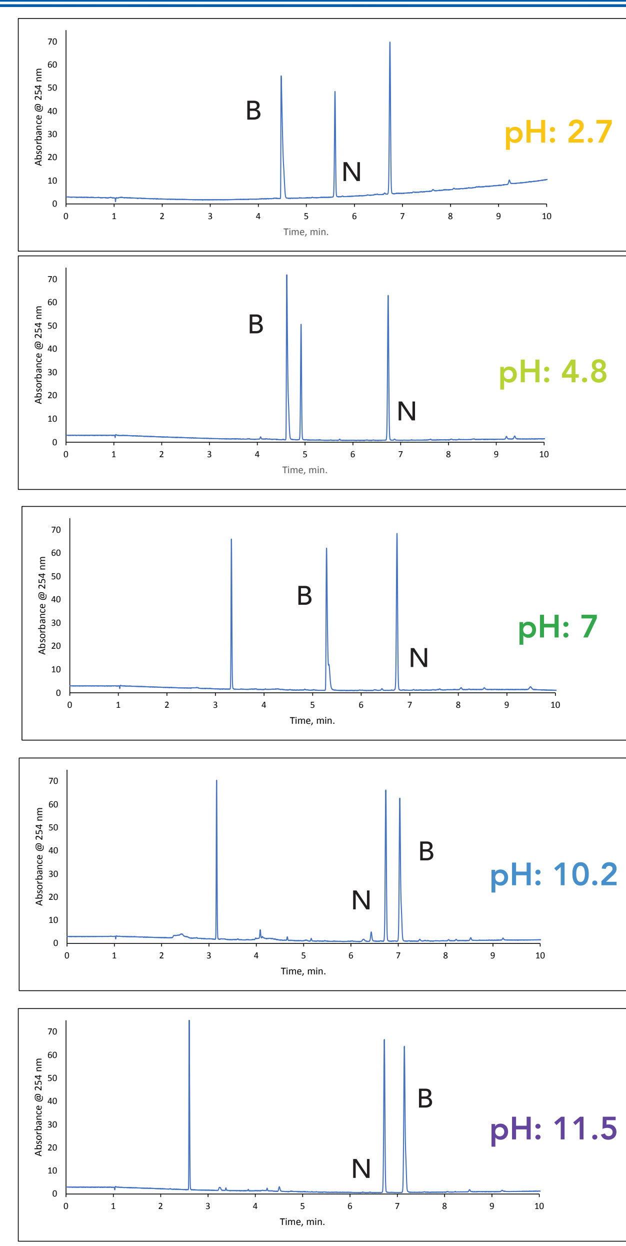
Adjusting the pH of the HPLC mobile phase is a great way to alter selectivity, particularly for ionizable compounds such as acids and bases. Depending on the analyte of interest, changing the pH of the mobile phase can also give retention time improvements along with significant peak shape advantages.

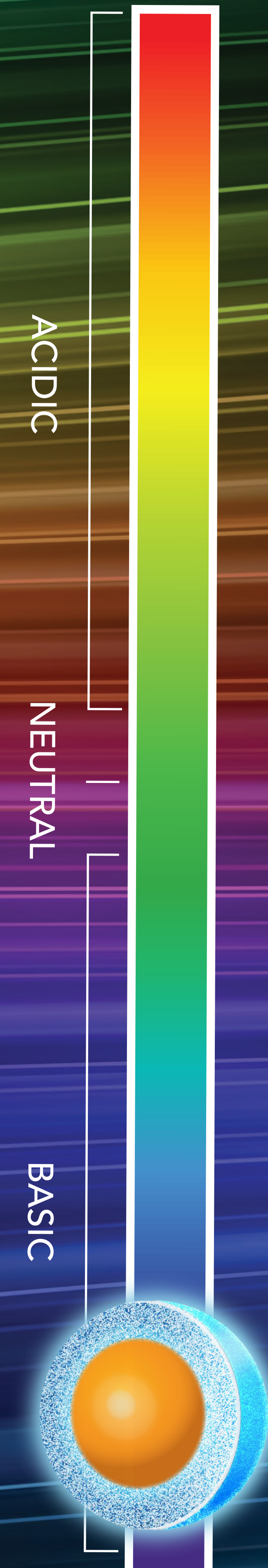
The HALO® ELEVATE column line incorporates a wide pH compatibility which allows not only chromatographic separations to be performed under low pH conditions, but high pH options are also available. (pH 2-12) This versatility allows for easier method development along with achieving selectivity differences that may benefit separation. This figure shows a mixture of an acid, base, and neutral analyte ran under low, medium, and higH pH conditions. *All HALO® ELEVATE phases demonstrate positive performance across the range...*

HALO® ELEVATE PHENYL-HEXYL

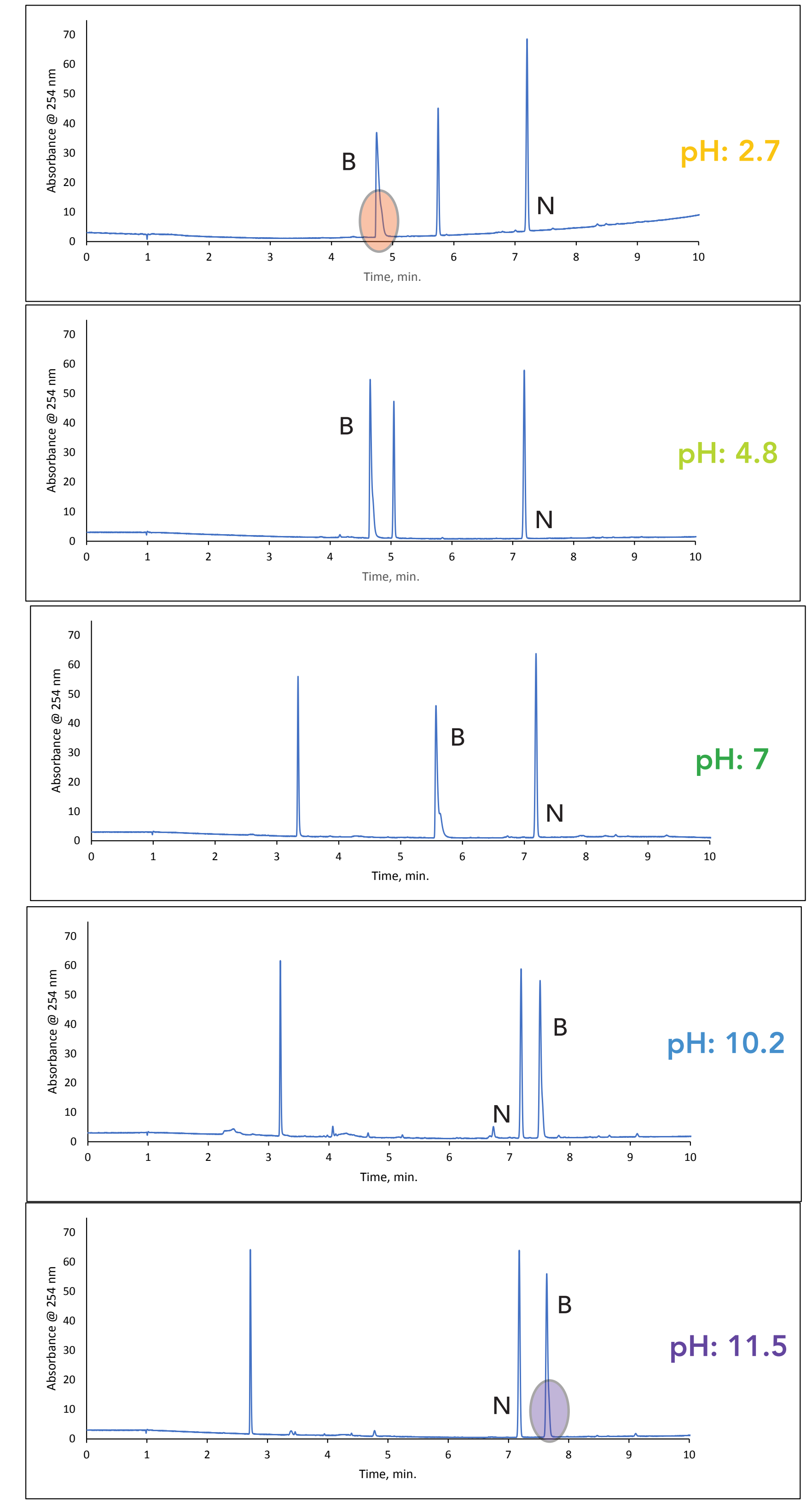


HALO® ELEVATE C8





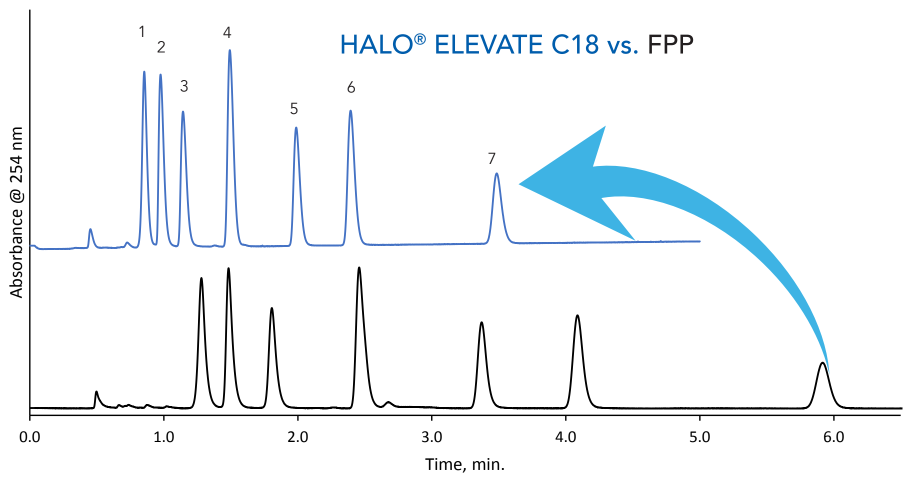
SYMMETRICAL PEAK SHAPE FOR THE BASIC COMPOUND IS ACHIEVED AT HIGH PH USING HALO® ELEVATE C18



For mobile phase conditions, contact us at halocolumns.com

ANTIDEPRESSANTS

A separation of antidepressants is achieved under high pH conditions using a HALO® ELEVATE C18 and Phenyl-Hexyl column against the FPP competitor columns. The separations using HALO® were completed in approximately half the time with improved resolution.



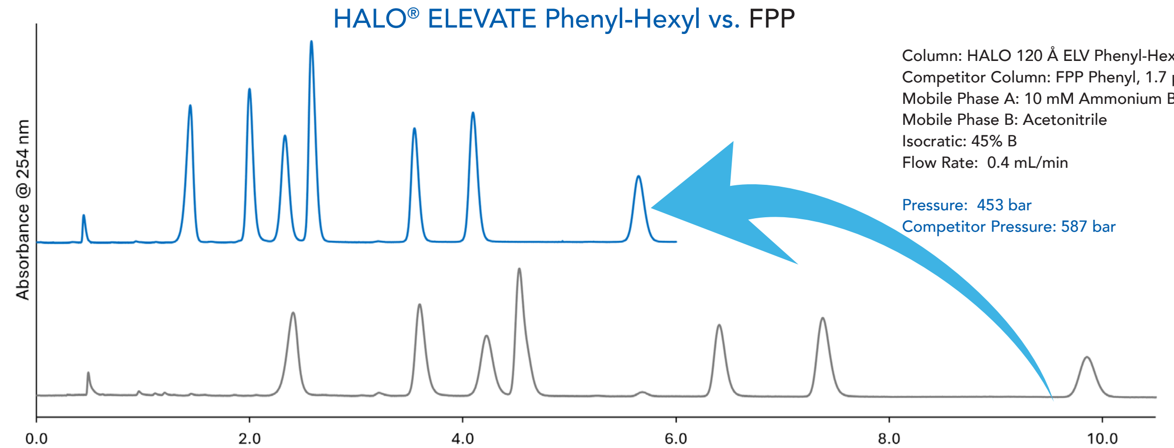
PEAK IDENTITIES

1. Amoxapine
2. Desipramine
3. Nortriptyline
4. Doxepin
5. Imipramine
6. Amitriptyline
7. Trimipramine

TEST CONDITIONS

Column: HALO 120 Å ELV C18 , 2 µm, 2.1 x 100 mm
Competitor Column: FPP C18, 1.7 µm, 2.1x100 mm
Mobile Phase A: 10 mM Ammonium Bicarbonate, pH: 10
Mobile Phase B: Acetonitrile
Isocratic: 60% B
Flow Rate: 0.4 mL/min

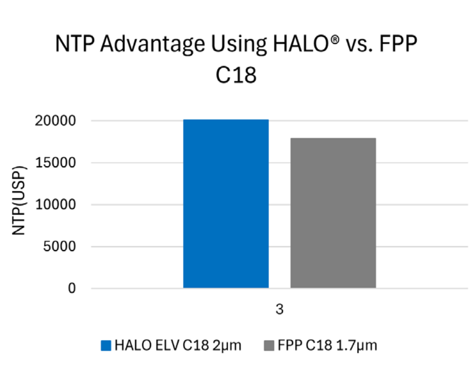
Pressure: 416 bar
Competitor Pressure: 519 bar

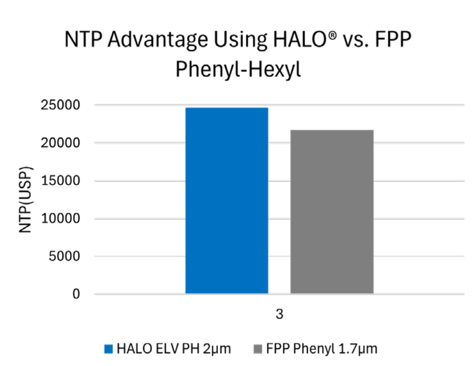


Column: HALO 120 Å ELV Phenyl-Hexyl , 2 µm, 2.1 x 100 mm
Competitor Column: FPP Phenyl, 1.7 µm, 2.1x100 mm
Mobile Phase A: 10 mM Ammonium Bicarbonate, pH: 10
Mobile Phase B: Acetonitrile
Isocratic: 45% B
Flow Rate: 0.4 mL/min

Pressure: 453 bar
Competitor Pressure: 587 bar

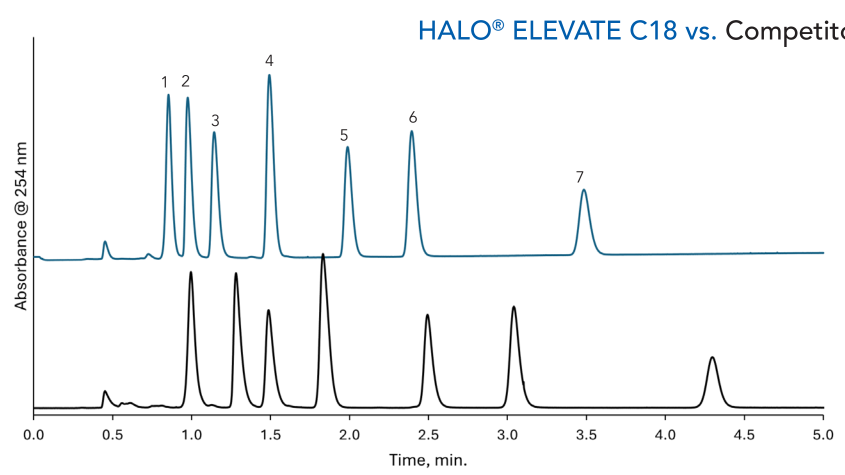
Increased surface area of smaller particle size enhances interactions between the stationary phase and analytes, producing sharper peaks and improved compound separation. This is reflected in higher theoretical plate counts as shown to the right in the bar chart where a 2µm HALO® was run against the similar FPP sized particle. (peak #7 from above is represented in bar chart).





HALO® ELEVATE C18 VS. COMPETITOR SPP C18 AT HIGH PH

A separation of antidepressants is achieved under high pH conditions using a HALO® ELEVATE C18 compared to a competitor high pH stable SPP column. Utilizing the same conditions the HALO® column acheives comparable resolution in a faster run time.



PEAK IDENTITIES

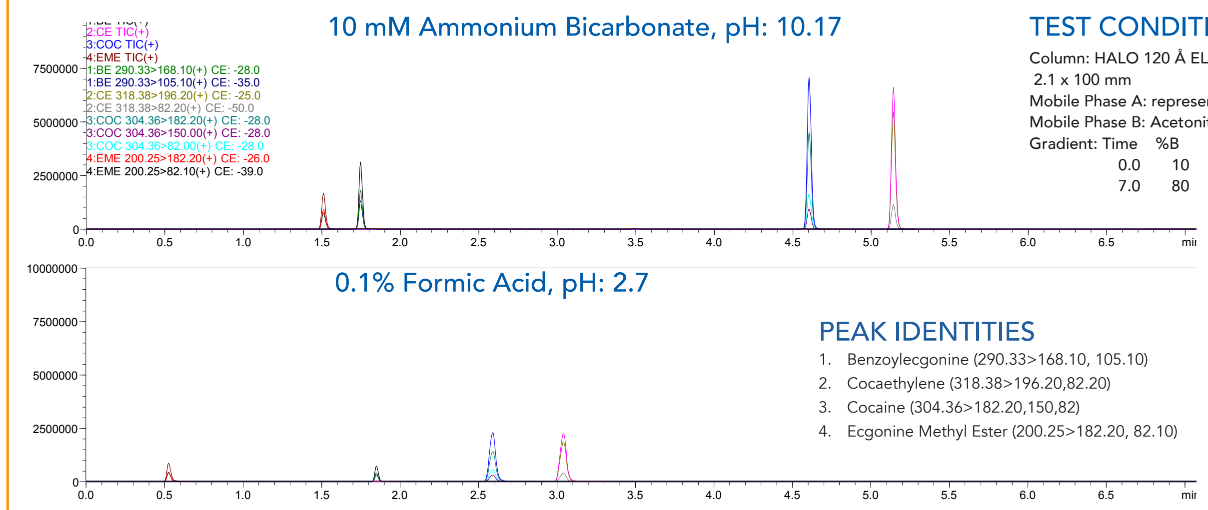
1. Amoxapine
2. Desipramine
3. Nortriptyline
4. Doxepin
5. Imipramine
6. Amitriptyline
7. Trimipramine

TEST CONDITIONS

Column: HALO 120 Å ELV C18 , 2.7 µm, 2.1 x 100 mm
Competitor Column: COMPETITOR SPP
Mobile Phase A: 10 mM Ammonium Bicarbonate, pH: 10
Mobile Phase B: Acetonitrile
Isocratic: 60% B
Flow Rate: 0.4 mL/min
Pressure: 205 bar
Competitor Pressure: 237 bar
Temperature: 35 °C

IMPROVED LC-MS PEAK SHAPES AT HIGH PH

Low pH/acidic mobile phases are typically used to separate cocaine (base) and its metabolites because cocaine is protonated and leads to high ionization/ sensitivity in ESI(+). High pH mobile phase/basic mobile phases tend to lead to better chromatography for basic analytes, leading to increased retention and improved peak shape.
For the best results, mobile phase pH should be above at least 1 pka unit when analyzing basic compounds under high pH conditions.



TEST CONDITIONS

Column: HALO 120 Å ELEVATE C8, 2.7 µm, 2.1 x 100 mm
Mobile Phase A: represented in chromatogram
Mobile Phase B: Acetonitrile

Gradient: Time	%B
0.0	10
7.0	80

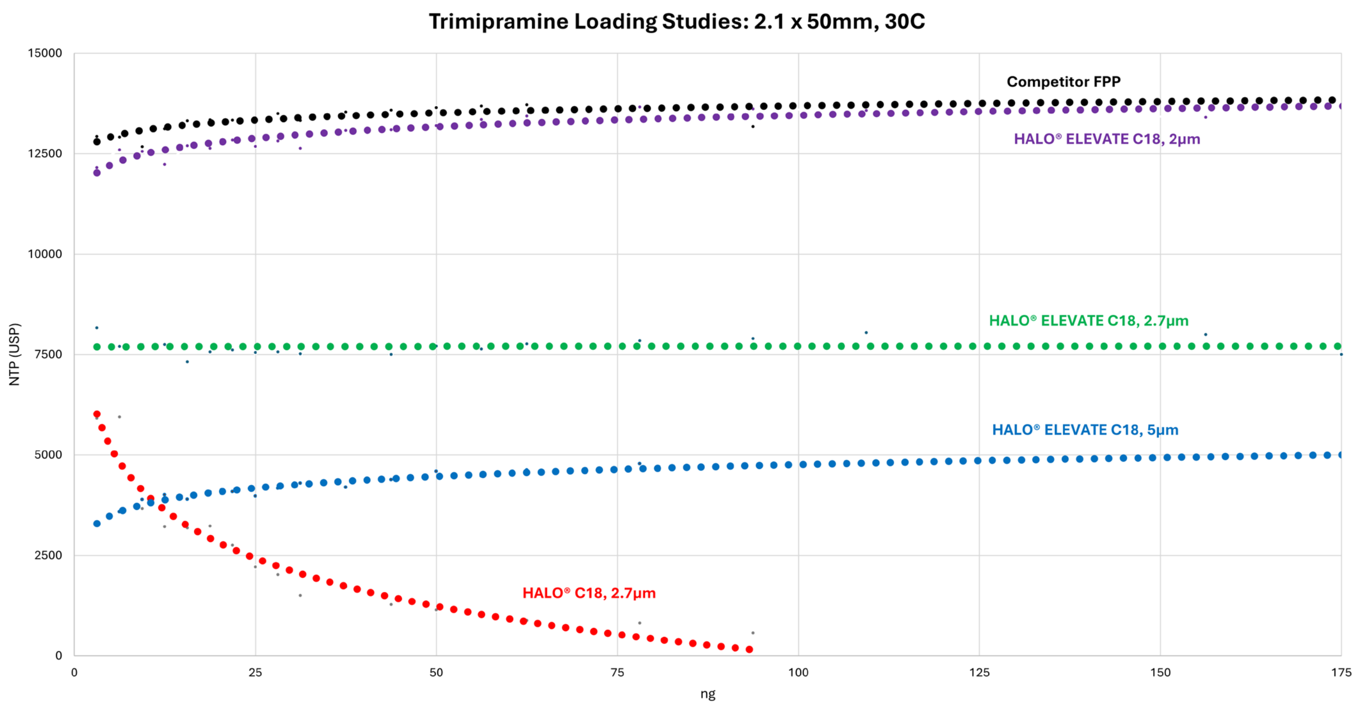
Flow Rate: 0.4 mL/min
Back Pressure: 265 bar
Temperature: 30 °C
Injection Volume: 0.1 µL

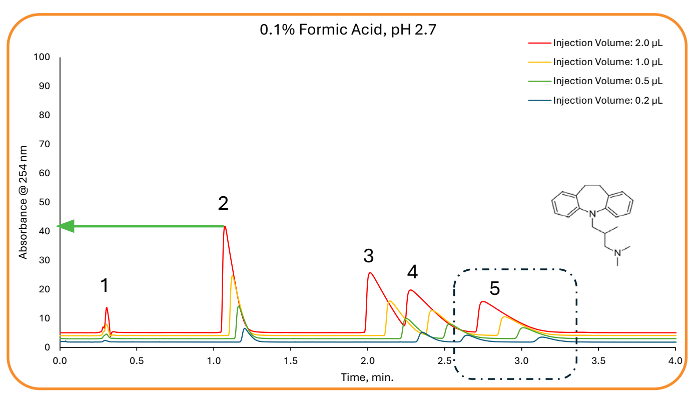
PEAK IDENTITIES

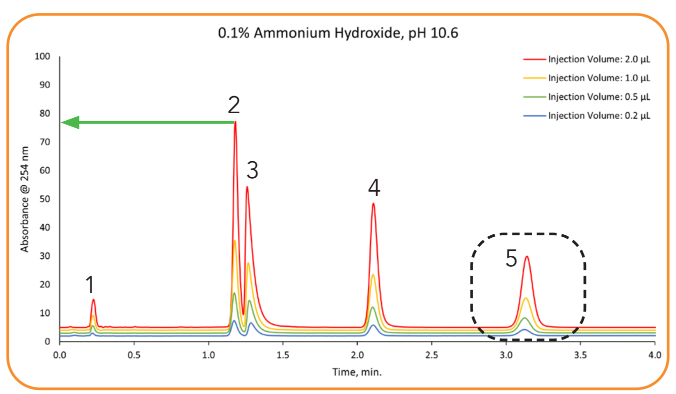
1. Benzoylecgonine (290.33>168.10, 105.10)
2. Cocaethylene (318.38>196.20,82.20)
3. Cocaine (304.36>182.20,150,82)
4. Ecgonine Methyl Ester (200.25>182.20, 82.10)

TRIMIPRAMINE LOADING STUDIES- LOW VS. HIGH PH

At low pH, basic compounds become positively charged which allow for unwanted interactions between the stationary phase/ silanols on the silica surface. Under high pH conditions, basic molecules become deprotonated, increasing retention (becoming less polar) and significantly improving chromatographic peak shape/ efficiency. This allows for much higher sample loading capacities compared to low pH conditions as seen in the figure below. In general, the pH of the mobile phase should be set to 2 units above the analyte's pKa value, allowing the base to become deprotonated (neutral). Below demonstrates trimipramine, a common tricyclic antidepressant (base) observed under low and high pH conditions at different sample loads on column.

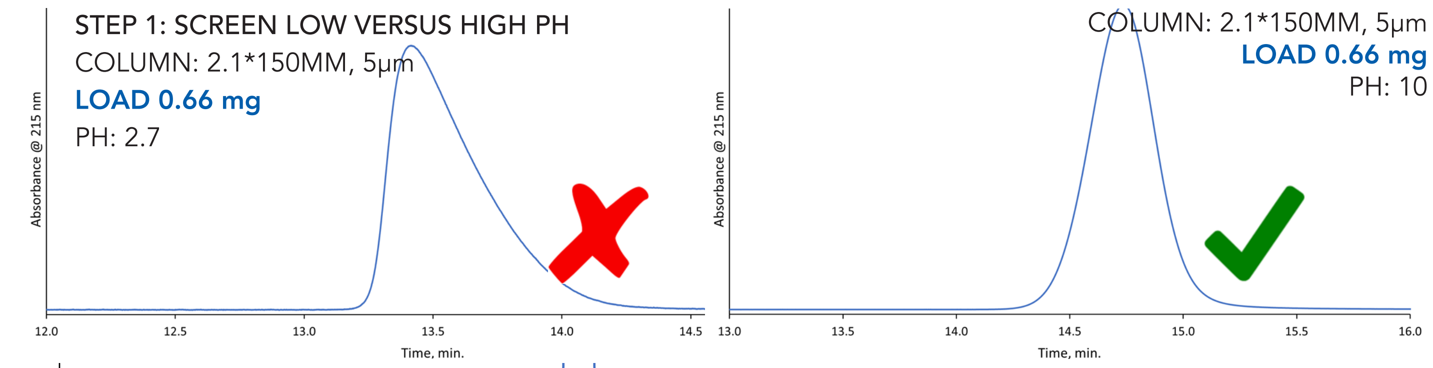


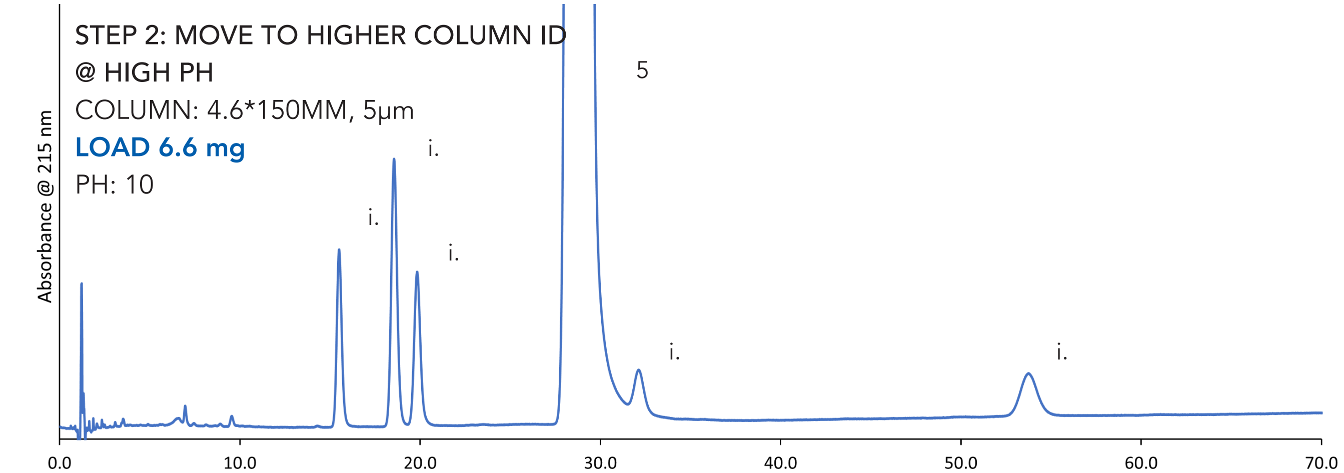


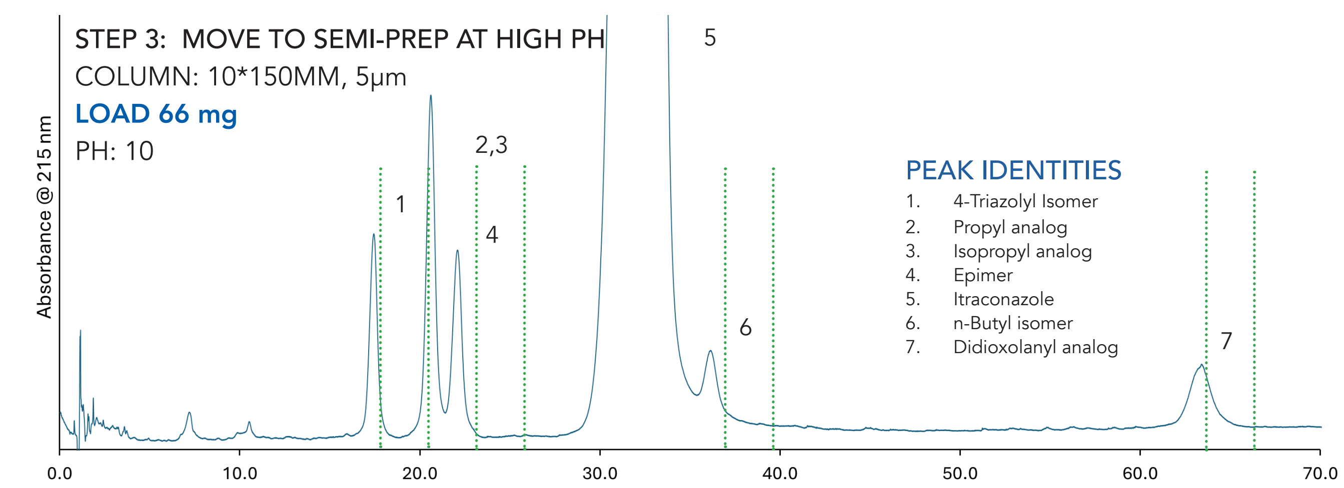


ITRACONAZOLE IMPURITY ANALYSIS VIA SEMI-PREP

Below demonstrates a separation of itraconazole and related impurities under low pH and high pH testing conditions. Peak shape is significantly improved under alkaline conditions, allowing for higher sensitivity for related impurities. As you increase the internal diameter of a column, you are also increasing the total loading capacity, allowing a larger amount of sample to be injected on column. In this test using the C18 phase, the 2.1 mm ID column was then scaled up to a 4.6mm ID, followed by a 10mm semi-preparative HALO® ELEVATE column effectivly loading 100x the sample on column.







With the higher sample load, impurities are identified, then fraction collected, dried down, and injected on a mass spectrometer to identify the impurities mass.

HALO® ELEVATE COLUMN PART NUMBERS

	2.0 µm PARTICLE SIZE			2.7 µm PARTICLE SIZE		
ID X L	C18	C8	PhHx	C18	C8	PhHx
1.5x50	9127X-402	9127X-408	9127X-406	9227X-402	9227X-408	9227X-406
1.5x100	9127X-602	9127X-608	9127X-606	9227X-602	9227X-608	9227X-606
1.5x150	9127X-702	9127X-708	9127X-706	9227X-702	9227X-708	9227X-706
2.1x20	91272-302	91272-308	91272-306	92272-302	92272-308	92272-306
2.1x30	91272-302	91272-308	91272-306	92272-302	92272-308	92272-306
2.1x50	91272-402	91272-408	91272-406	92272-402	92272-408	92272-406
2.1x100	91272-602	91272-608	91272-606	92272-602	92272-608	92272-606
2.1x150	91272-702	91272-708	91272-706	92272-702	92272-708	92272-706
2.1x250				92272-902	92272-908	92272-906
3.0x30	91273-302	91273-308	91273-306	92273-302	92273-308	92273-306
3.0x50	91273-402	91273-408	91273-406	92273-402	92273-408	92273-406
3.0x100	91273-602	91273-608	91273-606	92273-602	92273-608	92273-606
3.0x150	91273-702	91273-708	91273-706	92273-702	92273-708	92273-706
3.0x250				92273-902	92273-908	92273-906
4.6x30				92274-302	92274-308	92274-306
4.6x50				92274-402	92274-408	92274-406
4.6x100				92274-602	92274-608	92274-606
4.6x150				92274-702	92274-708	92274-706
4.6x250				92274-902	92274-908	92274-906
10x150				92270-702	92270-708	92270-706
20x150				9227A-702	9227A-708	9227A-706
Guard (3/pk)	91272-102	91272-108	91272-106	92272-102	92272-108	92272-106
Guard (3/pk)	91273-102	91273-108	91273-106	92273-102	92273-108	92273-106
Guard (3/pk)				92274-102	92274-108	92274-106
Guard Holder	94900-001					

	5 µm PARTICLE SIZE		
IDxL	C18	C8	PhHx
2.1x30	95272-302	95272-308	95272-306
2.1x50	95272-402	95272-408	95272-406
2.1x100	95272-602	95272-608	95272-606
2.1x150	95272-702	95272-708	95272-706
2.1x250	95272-902	95272-908	95272-906
3.0x30			
3.0x50			
3.0x100			
3.0x150			
3.0x250			
4.6x30			
4.6x50	95274-402	95274-408	95274-406
4.6x100	95274-602	95274-608	95274-606
4.6x150	95274-702	95274-708	95274-706
4.6x250	95274-902	95274-908	95274-906
10x150	95270-702	95270-708	95270-706
20x150	9527A-702	9527A-708	9527A-706
Guard(3/pk)	95274-102	95274-108	95274-106
Guard Holder	94900-001		

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