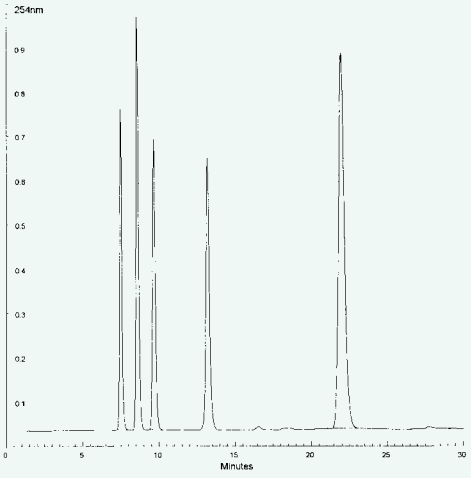
Higgins Analytical

HAISIL 300 C18 12µm Coupled to ☞ HAISIL 100 C18 5µm
Dimensions 25 x 20mm
Catalog No. HF-0220-W182
Batch No. 260402

Dimensions 250 x 20mm
Catalog No. HS-2520-M185



Catalog No.	HF-0220-W182			
Description:	HAISIL 300 C18 12µm			
Dimensions:	25 x 20mm			
Sorbent Lot#:	P25076	Monofunctional/Endcapped		
Particle Size:	12µm			
Pore Size:	300 Å			
Mobile Phase:	70% Acetonitrile in Water			
Flow Rate:	10mL/min			
Pressure:	bar		Temp:	28.5 °C
Peak:	tR(min)	35 bar	Plates(N)	Skewness
# 1	7.50	Dimethyl Phthalate	10,655	0.328
# 2	8.59	Nitrobenzene	10,725	0.298
# 3	9.67	Anisole	11,094	0.294
# 4	13.19	Diphenylamine		
# 5	22.01	Fluorene	12,886	0.238

File: 172 26/4

HAI*speed* 20 Guard Cartridges *(continued):*

Uncompromising Performance:

Volume variance calculations for the data illustrated on the previous page show that the 25mm cartridge has approximately 500 theoretical plates; not bad for a 2.5cm long column packed with 10 - 15µm particles! And, more importantly, the coupled guard and prep column assembly has better performance than most manufacturer's prep columns without a guard.

0.5µm Frits for Maximum Protection:

Another unique feature of HAI*speed* 20 guard cartridges is the 0.5µm inlet and outlet frit. We've built in-line filtration right into the cartridge!

HAI*speed* 20 Prep Cartridges are shipped with end caps that prevent solvent loss and protects the cartridge and frit.

High Speed Microbore Analysis *Application Highlights:*

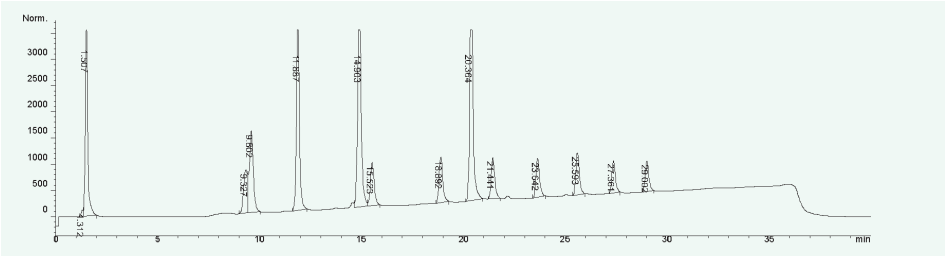
- High stability and low back pressure (150bar) even at 6 times the optimum flow rate.
- Basic drugs don't tail on TARGA columns when using only a mild formic acid buffer that is ideal for LC/MS applications.
- The TARGA column shows unique selectivity, performs well, and equilibrates quickly in 100% aqueous conditions.

High Speed Microbore Analysis of Plasma Drug Samples:

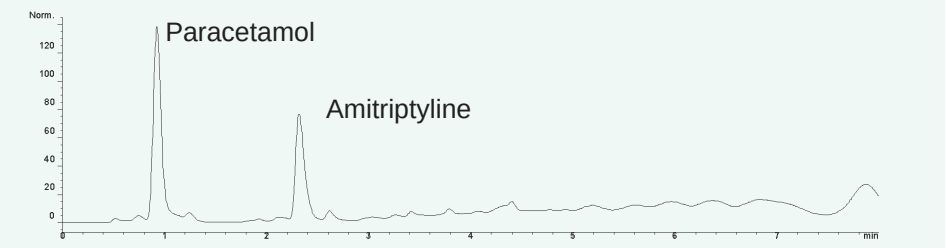
Column stability and efficiency are primary issues for researchers developing fast HPLC methods with low detection limits. An example of a very high throughput plasma drug analysis illustrates several performance features of Higgins Analytical's TARGA microbore columns.

Experimental: 200µL of plasma is precipitated with MeCN, evaporated to dryness, then dissolved in 10µL of 10% MeOH/water solution. The 2.5µL injection on a TARGA C18 100x1mm column is equivalent to 50µL of plasma. A 0 - 95% MeCN vs 0.1% aqueous formic acid gradient is run at 300µL/min over 5min, held for 1min, then returned to 0% MeCN by 6.1min. The analysis is repeated every 8 minutes.

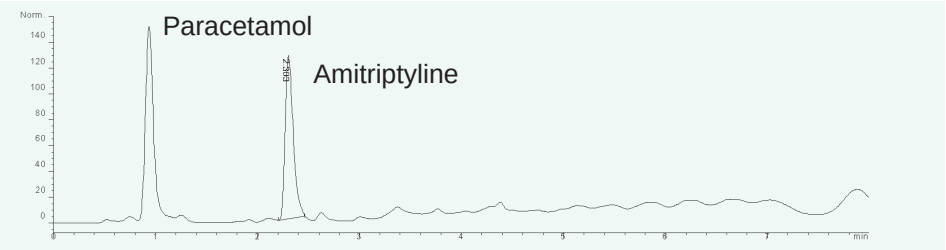
Good column efficiency and lifetime are characteristics of TARGA C18 columns, even under these demanding conditions.



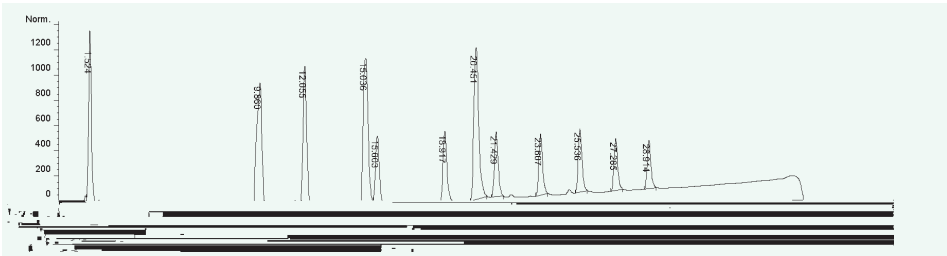
A thirteen component *Column Suitability Gradient Test* on a new 100x1.0mm TARGA C18 Column, P/N TS-1001-C185.



The first analysis of a 10µg/mL plasma drug sample on a new 100x1.0mm TARGA C18 Column. Note the good amitriptyline peak symmetry and efficiency at 300µL/min.



The 80th analysis of plasma drug samples on a 100x1.0mm TARGA C18 Column. No change in retention times or peak shapes even under the high flow rate conditions.

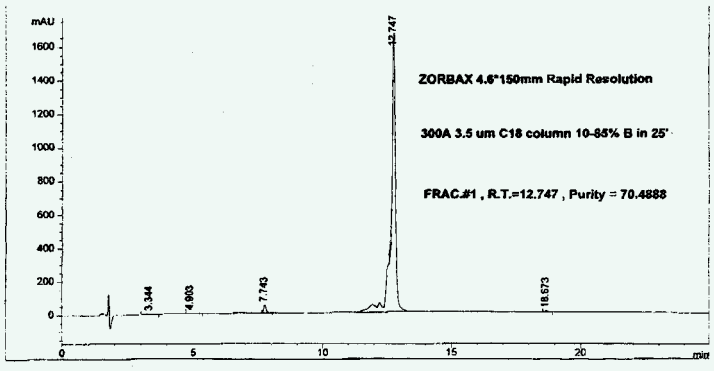


A repeat of the thirteen component *Column Suitability Test* on a 100x1.0mm TARGA C18 Column after 80 analysis of plasma drug samples at a flow rate of 300 µL/min.

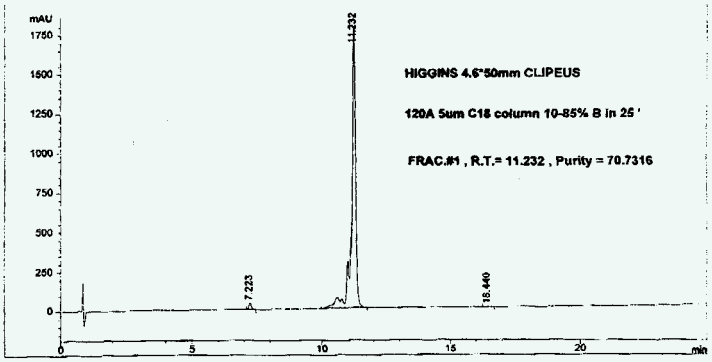
Higgins Analytical is grateful to Rob Plumb, BioMet, Glaxo Wellcome, Hertfordshire, UK for permission to share these TARGA 1mm data.

CLIPEUS selectivity wins another horsepower race:

Particle size, pore size, column length, or selectivity - which is the most important parameter to consider when optimizing a chromatographic separation?



Analysis of a preparative chromatography fraction on a Zorbax 3.5µm Rapid Resolution Column, 150x4.6mm.



Repeat analysis of the preparative chromatography fraction illustrated on top using a CLIPEUS 5µm C18 Column, 50x4.6mm.

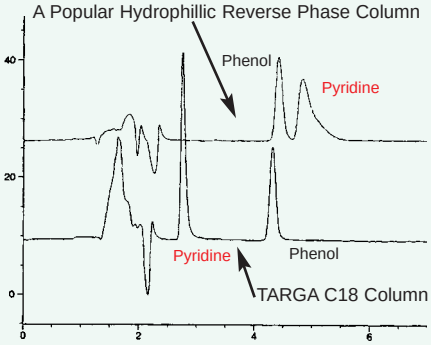
Unfortunately, the sequence listed above is what many analysts follow when developing new, or optimizing old, HPLC methods. Realizing that selectivity is the most powerful factor, an experienced chromatographer approaches the challenge by considering these four factors in the reverse order.

Preparative chromatography fractions of a crude synthetic peptide were analyzed on a Zorbax 150x4.6mm 3.5µm 300Å Rapid Resolution C18 column and a CLIPEUS 50x4.6mm 5µm 120Å C18 column. The results in the chromatograms remind us that column length plays a minor role in gradient separations and illustrates that the selectivity and increased surface area of the CLIPEUS column out-performs a column three times longer and packed with particles 2/3 the diameter. Lower cost, lower operating pressure, and lower risk of clogging are additional advantages the shorter, larger particle size CLIPEUS column offers.

In addition to the C18 phase used in this example, the CLIPEUS columns family includes C8, Cyano, and Phenyl phases. These columns and the derivation of the CLIPEUS name are described in our new ***1999 - 2000 Product Catalog*** available from your local distributor.

Using TARGA for analysis of polar solutes in high aqueous conditions:

Since their introduction in 1996, TARGA HPLC columns and cartridges have become a popular choice for LC-MS applications, especially ones involving problematic basic compounds. The inert surface chemistry characteristics and relatively high surface area provide a unique selectivity without peak tailing even for basic samples. More importantly, problematic solutes such as basic peptides can be analyzed without TFA in the mobile phase which greatly enhances detection limits. Buffers as weak as 0.01% acetic or formic acid are routinely used for peptide mapping and peptide analysis.



TARGA C18 compared to a popular column designed for working with high aqueous mobile phases. 150X4.6mm, 20% MeCN/water (unbuffered), 1mL/min.

TARGA HPLC columns work well with 100% aqueous mobile phases and equilibrate quickly when running gradients beginning with 100% water (see the paracetamol/amitriptyline analysis on the opposite page).

Wide Range for Formats Offered:

Higgins Analytical products are available in cartridge as well as conventional column formats. Our small 20x2.1mm cartridge columns can be used as column guards or as efficient analytical columns for high speed and LC-MS applications.

Combinatorial Chemistry.... Why 2µm columns will not address this new technology's high throughput requirements.

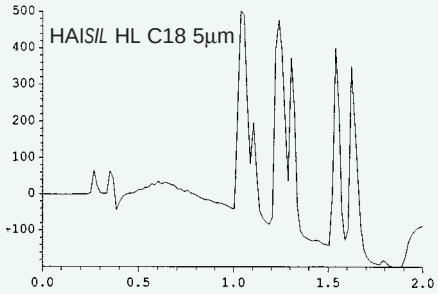
Combinatorial Chemistry is another example of an emerging technology that is going to place new demands on HPLC for speed and assay robustness.

Industry innovators continue to introduce "new" technology HPLC columns based on non-porous and porous particles with 2µm or smaller diameters. There have even been rumblings about columns packed with 1µm particles. Don't hold your breath! The physical constraints of column diameter, particle size, and column length were described over 20 years ago, but we have a penchant for ignoring these teachings as we get swept away by today's marketing enthusiasm.

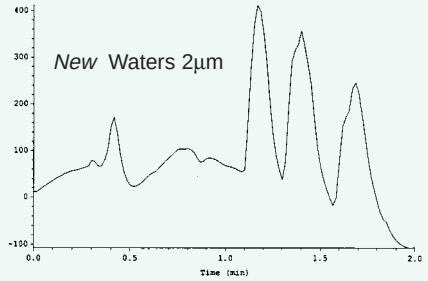
The following chromatograms illustrate how much more important the roles of selectivity and good column packing technology are in comparison to particle size when it comes to sheer horsepower in HPLC separations.

This important point is illustrated by the analysis of two combinatorial libraries on two manufacturers' 2µm columns and comparing the results of the same analysis on a short HAISIL HL C18 5µm column.

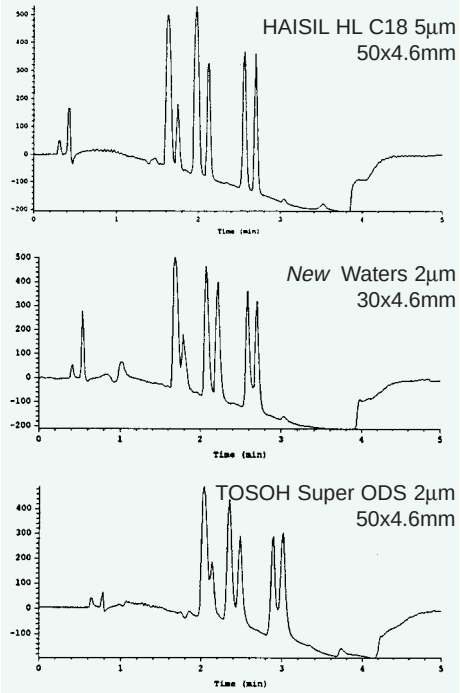
The objective of this study is to resolve the two multicomponent mixtures with a fast gradient in as little as two minutes. In each case the 5µm HAISIL HL C18 column clearly out performed the 2µm alternatives.



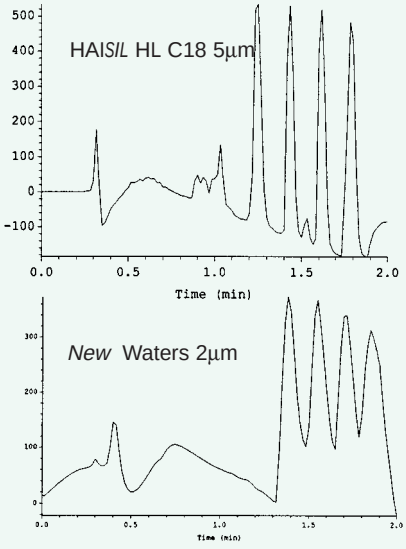
A six component test mix analyzed on a 50x4.6 mm HAISIL HL C18 5µm column with a fast MeCN/water 0.1% TFA gradient.



The same six component test mix analyzed on a *new* Waters 30x4.5mm 2µm combinatorial analysis column.



The same six component library shown at the top of the column to the left analyzed with a slower MeCN/water 0.1% TFA gradient.



A four component library analyzed with a fast gradient on a HAISIL HL C18 5µm (top) and new Waters 30x4.6mm 2µm column (bottom).

Find out more about these and other Higgins Analytical products in the new Catalog available from your local distributor.

Higgins Analytical Products are sold and supported by distributors worldwide:

Higgins Analytical, Inc.

Expert Manufacturer of HPLC Consumables

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