the RESTEK ADVANTAGE

2006.02

Allure Biphenyl™ HPLC Columns

Exclusive to Restek!

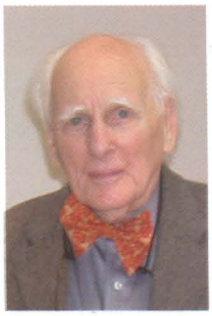
- Unique stationary phase promotes π-π interactions with aromatic and unsaturated compounds.
- Enhanced selectivity for steroids, contraceptive hormones, tetracyclines, explosives.
- Improved retention, relative to traditional phenyl phases, for unsaturated or saturated compounds.

See **page 4**.

IN THIS ISSUE

Erratum
Exempted drugs of abuse reference materials alprazolam (cat.# 34042), chlordiazepoxide HCl (cat.# 34044) and levorphanol (cat.# 34003) are at a concentration of 1mg/mL, not as listed in Advantage 2006v1.

Daily drawing winners at our PittCon® booth
Monday: Dr. S. Todd Swanson, University of Nebraska, Lincoln, NE
Tuesday: Dr. Jeffery Loo, General Motors, Milford, MI
Wednesday: Dr. Steven DuBose, Alcon Research, Fort Worth, TX
Thursday: Dr. Shawn Shanmugan, US Smokeless Tobacco, Nashville, TN
Congratulations, gentlemen, and thank you to everyone who visited our booth!



By Professor Walter Jennings ("Walt") waltj@pacbell.net
Professor Emeritus, University of California, Davis
Co-Founder, J&W Scientific, Inc.
Co-Founder, AirToxics, Ltd.

Preventive Maintenance for GC

Far too many practitioners install a new column, then ignore the enclosures that came with that column, and blissfully proceed to analysis. For reasons discussed below, most authorities would agree that the "typical" or "generic" test chromatograms that accompany batch-tested columns are essentially useless and can be ignored, but if the enclosures include a test chromatogram *that is specific for that column*, this can be valuable. The user gains a distinct advantage by purchasing individually tested columns. In batch testing, one or more columns are tested and regarded as representative of the quality of that entire batch. But the quality of a given batch of columns - - whether N, N/m, bleed, or level of response to active analytes - - normally follows a Gaussian distribution, and individual testing allows the discriminating manufacturer to identify and discard those columns on the low end of any given quality curve. With batch-tested columns, those sub-standard columns become part of the stock shipped to customers. The individually tested column offers another advantage: the test chromatogram is specific for that column under a set of specified conditions. Manufacturers using the more expensive procedures of individual testing must maintain the QC testing chromatographs in pristine condition. Chromatographs possessing even trace residues in the injectors, detectors, or gas supply lines, or faulty temperature readouts can result in the condemnation of columns that are actually good. To prevent this expensive blunder, injectors, detectors, and gas supply lines must be routinely cleaned and/or replaced; in addition, oven readouts (which do drift) require periodic recalibration, and some manufacturers do this on a weekly basis.[1]

The test chromatogram for an individually tested column illustrates the chromatogram produced by a specific test sample under specified conditions in a meticulously maintained instrument. After installing a new column, it should be conditioned in accordance with the manufacturer's recommendations. One should then make an injection of that same test mixture under the same conditions and compare the results to the test chromatogram. *Differences in theoretical plate numbers, the relative responses of test solutes, retention factors or separation factors may indicate instrumental problems that should not be ignored.*

Brandies and some wines improve with age, but most other things undergo a time-related deterioration. Neither gas chromatographs (nor unfortunately, the author) are exceptions to this generality. Eventually problems invariably emerge - - for the GC, these can take the form of unsteady baselines, erratic signal, noise spikes, ghost peaks, higher detection limits, and higher bleed. Some users become addicted to a short-term solution: they simply install a new column and the problem disappears - - for a time. This solution is especially common for those analysts under pressure to produce results rapidly because more samples are coming in the door. However, column replacement is but a temporary solution, in that the same semi-volatile contaminants that destroyed the last column are now being trapped on the new column. Until they work their way through the column and to the detector, the analyst is lulled. However, eventually they do reach the detector, and the problem recurs. In the absence of corrective actions, the interval between the need for replacement columns continues to become shorter and shorter. This dilemma is exacerbated by "real world" or dirty samples, but it also occurs, albeit less frequently, for those analyzing pristine samples.

Where do these problems originate? Few samples are truly clean. It can be educational to place a few mL of the sample on a clean watch glass, allow it to evaporate, and note the residue. In addition to these semi-volatile and non-volatile sample residues, we should be concerned with gas-borne contaminants - - with FID, these would include carrier, combustion hydrogen, make-up, and air. Most chromatographers recognize that there are different purity grades for gases, and the wary analyst specifies "five-nines-purity" (99.999%). This does not negate the need for gas filters (or traps), but usually ensures that the filters will last longer. Removal of oxygen from the gas streams is important. Even traces of oxygen attack the siloxane chain (on which most GC polymers are based), cleave Si-C bonds, and leave terminal Si-OH groups at the points of cleavage. This quickly converts the column into a "bleeder", because the terminal silanols encourage "back-biting" reactions. These split out cyclic siloxanes, primarily trimers and tetramers of (-Si-O-), generating new terminal Si-OH groups at the points of cleavage. Oxygen traps normally pay for themselves because columns experience longer lifetimes. Water traps are also important, not for the stationary phase per se, but because water or water vapor will cause most oxygen scrubbers to deteriorate rapidly. Hence good judgment dictates that the carrier should be passed first through a water trap, then through a bulk oxygen trap, and finally through an indicating oxygen trap. Traps should be mounted vertically, never horizontally. Most are filled with particulate materials and in the horizontal position the particles can settle, leaving an overhead void that provides a path of lower resistance through which most of the gas will flow.

By comparing the performance of a replacement column with the test chromatogram specific for that column, paying attention to sample composition and cleanliness, conducting proper injector and detector maintenance, and using high quality gas purifiers even with high purity gases, column lifetimes can be extended, and down times become a rarity. A regular schedule of preventive maintenance does pay dividends.

[1] Temperature exercises an exponential effect on solute retention factors (k). One of the more precise methods of re-setting the oven readout is to reserve, solely for that purpose, a "recalibration column" on which solute k values at known temperatures have been predetermined. Temperature controls are manipulated to produce the proper solute retention factors, and the readout reset

Inlet Seals for Agilent Instruments

by Donna Lidgett, GC Accessories Product Marketing Manager

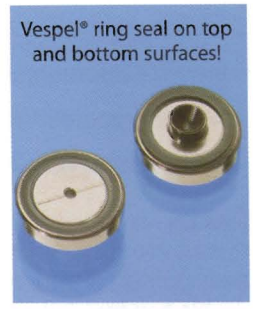
Dual Vespel® Ring Inlet Seals

- Vespel® ring embedded in bottom surface eliminates need for washer.
- Vespel® ring embedded in top surface reduces operator variability by requiring minimal torque to seal.
- Prevents oxygen from entering the carrier gas, increasing column lifetime.

In Agilent split/splitless injection ports, our Dual Vespel® Ring Inlet Seal greatly improves performance, relative to conventional metal-to-metal seals—it stays sealed, even after repeated temperature cycles, without retightening the reducing nut! Two soft Vespel® rings, outside the sample flow path, eliminate the need for a washer and ensure very little torque is needed to make a leak-tight seal. Tests show Dual Vespel® Ring Inlet Seals seal equally effectively at torques from 5 in. lb. to 60 in. lb.

Use a stainless steel seal for analyses of unreactive compounds. To reduce breakdown and adsorption of active compounds, use a Siltek®-treated or gold-plated seal.

0.8mm ID Dual Vespel® Ring Inlet Seal	2-pk.	10-pk.
Siltek®	21242	21243
Gold-Plated	21240	21241
Stainless Steel	21238	21239
1.2mm ID Dual Vespel® Ring Inlet Seal	**2-pk.**	**10-pk.**
Siltek®	21248	21249
Gold-Plated	21246	21247
Stainless Steel	21244	21245



best choice!

Washerless, leak-tight seals for Agilent GCs

Patent pending.

Replacement Inlet Seals for Agilent 5890/6890/6850 Split/Splitless Injection Ports

- Special grade of stainless steel that is softer and deforms more easily, creating a better seal.
- Increases column lifetime because oxygen cannot permeate into the carrier gas.
- Reduced noise benefits high-sensitivity detectors (e.g., ECDs, MSDs).
- Siltek® treatment provides inertness similar to fused silica.
- All seals include washers.

The inlet seal at the base of the Agilent 5890/6890 GC injection port contacts the sample and, because septum fragments and sample residue accumulate on the seal surface, the seal must be changed frequently to prevent adsorption of active compounds.

Use a stainless steel seal for analyses of unreactive compounds. To reduce breakdown and adsorption of active compounds, use a Siltek®-treated or gold-plated seal.

Single-Column Installation, 0.8mm Opening*		0.25/0.32mm ID Dual-Column Installation, 1.2mm Opening		0.53mm ID Dual-Column Installation (1/16-inch opening)	
2-pk.	10-pk.	2-pk.	10-pk.	2-pk.	10-pk.
Stainless Steel Inlet Seal					
21315	21316	20390	20391	20392	20393
Gold-Plated Inlet Seal					
21317	21318	21305	21306	—	—
Siltek® Inlet Seal					
21319	21320	21307	21308	—	—

*0.8mm ID stainless steel inlet seal is similar to Agilent part #18740-20880, 0.8mm ID gold-plated inlet seal is similar to Agilent part #18740-20885.

tech tip

Use a 1.2mm inlet seal with Vespel®/graphite ferrules or when installing two columns using a two-hole ferrule. Use a 0.8mm inlet seal with graphite ferrules or single capillary column installations.

Replacement Inlet Seal Washers

Description	Similar to Agilent part #	qty.	cat.#
Replacement Inlet Seal Washers	5061-5869	15-pk.	21710

Optimizing Difficult Separations of Steroids

Using an Allure™ Biphenyl HPLC Column

By Rick Lake, Pharmaceutical Innovations Chemist

- Increase resolution while using simple, isocratic conditions.
- Achieve separations not possible on a C18 column.
- Rugged and reproducible analyses.

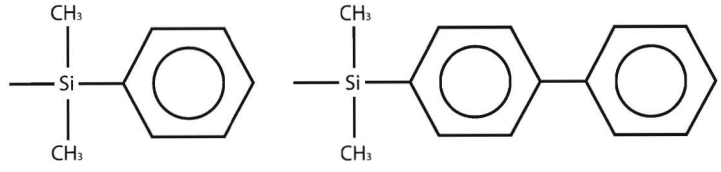
Steroids are an unusual class of compounds, in that all structural variation is centered on a common conjugated ring system, with differences in double bonding and ring constituents producing chemical diversity. Because of the consistency in their chemical structures, it can be difficult to achieve adequate separation of steroids on an alkyl (e.g., C18) HPLC stationary phase. An optimized stationary phase can be the key to these analyses.

When choosing a stationary phase, a separation mechanism that utilizes inherent differences in the chemical structures of the target analytes should be used. For analyses in which the target analytes are structurally very similar, this is especially critical. For steroids, this includes separations based on pi-pi (π-π) interactions between aromatic or unsaturated moieties: a stationary phase containing phenyl groups forms π-π bonds as the phenyl group on the stationary phase overlaps with the aromatic rings or double bonds in the analytes.

Restek chemists have made significant advancements in phenyl stationary phase chemistry, to increase retention of unsaturated compounds in reversed phase HPLC applications, while enhancing selectivity. The Allure™ Biphenyl stationary phase is a product of these advancements. A typical silica-based phenyl stationary phase consists of a single phenyl group bonded to a silica backbone (Figure 1). By developing a phase that consists of two phenyl groups bonded end-to-end, the Allure™ Biphenyl offers a more concentrated arrangement of phenyl groups, in a sterically favorable positioning (Figure 1). This phase shows markedly better selectivity for unsaturated compounds and shows a high retention capacity, similar to that of a C18 phase.[1-3]

We assayed two groups of steroids, hormones and corticosteroids, on an Allure™ Biphenyl column, to determine if separation can be enhanced by exploiting differences in π-π interactions. First, we compared performances by the Allure™ Biphenyl column and a conventional C18 column of the same dimensions, using a complex mix of steroid hormones. Under identical isocratic analytical conditions, the Allure™ Biphenyl column resolved all target compounds (Figure 2), but the C18 column showed very limited resolving power (Figure 3).

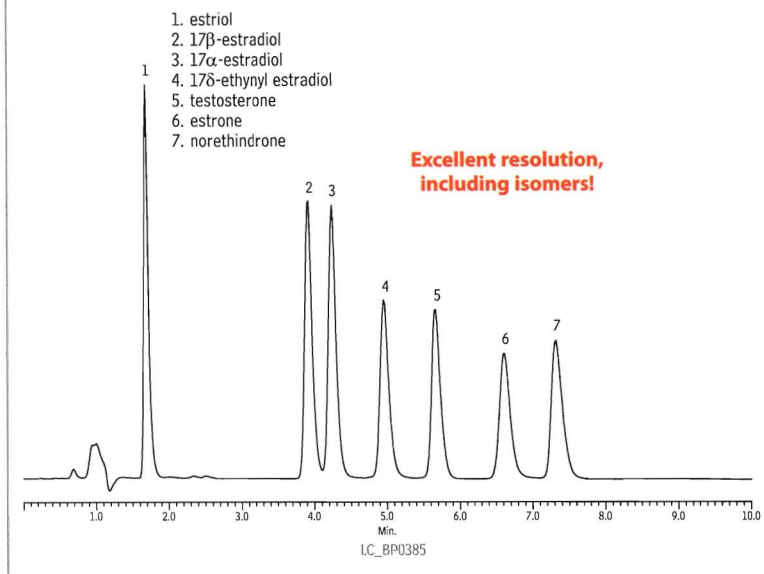
Figure 1 The unique chemistry of the Allure™ Biphenyl stationary phase creates a more concentrated phenyl arrangement in a sterically favorable positioning.



Conventional phenyl stationary phase

Allure™ Biphenyl stationary phase

Figure 2 An Allure™ Biphenyl column resolves steroid hormones in a simple, isocratic analysis.

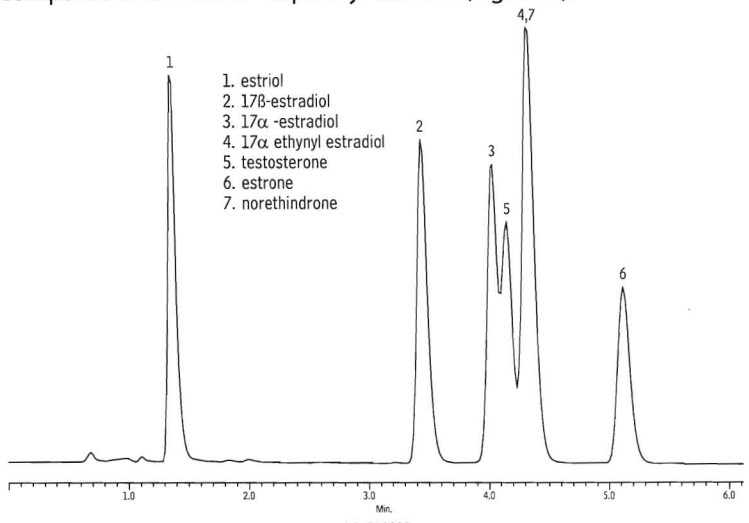


Sample:
Inj.: 10µL
Conc.: 50µg/mL each component
Sample diluent: acetonitrile:methanol, 4:1 (v/v)

Column: **Allure™ Biphenyl** (cat.# 5166365)
Dimensions: 150 x 4.6 mm
Particle size: 3µm
Pore size: 60Å

Conditions:
Mobile phase: water:acetonitrile, 50:50 (v/v)
Flow: 1.5mL/min.
Temp.: ambient
Det.: UV @ 220nm

Figure 3 A C18 column shows less resolving capability for steroids, compared to an Allure™ Biphenyl column (Figure 2).

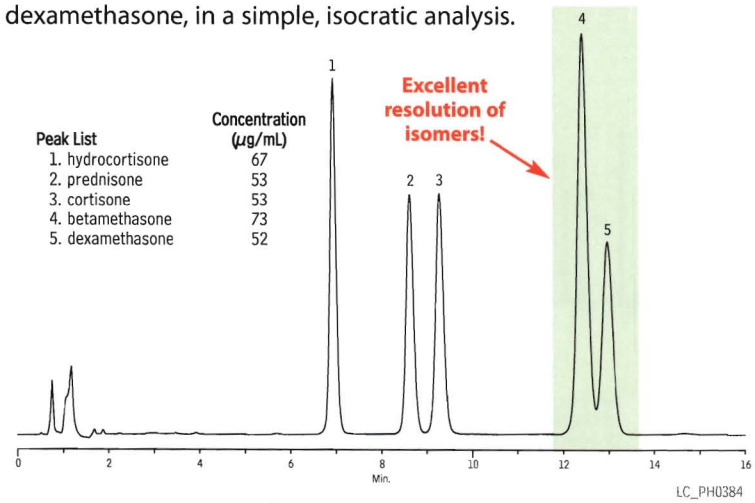


Sample:
Inj.: 10µL
Conc.: 50µg/mL each component
Sample diluent: acetonitrile:methanol, 4:1 (v/v)

Column: C18
Dimensions: 150 x 4.6 mm
Particle size: 3µm
Pore size: 100Å

Conditions:
Mobile phase: water:acetonitrile, 50:50 (v/v)
Flow: 1.5mL/min.
Temp.: ambient
Det.: UV @ 220nm

Figure 4 An Allure™ Biphenyl column resolves all target corticosteroids, including isomers betamethasone and dexamethasone, in a simple, isocratic analysis.



Sample:
Inj.: 10µL
Sample diluent: methanol

Column: Allure™ Biphenyl (cat.# 5166365)
Dimensions: 150 x 4.6 mm
Particle size: 3µm
Pore size: 60Å

Conditions:
Mobile phase: water:acetonitrile, 70:30 (v/v)
Flow: 1.5mL/min.
Temp.: ambient
Det.: UV @ 220nm

The Allure™ Biphenyl column also provided an overall increase in analyte retention – a very useful improvement relative to conventional phenyl phases.

The Allure™ Biphenyl column also showed enhanced selectivity in a second analysis, using corticosteroids. Under simple isocratic conditions, the Allure™ Biphenyl column provided baseline separation of hydrocortisone and prednisone and, more important, resolved isomers betamethasone and dexamethasone (Figure 4).

These analyses show that markedly better selectivity for steroids easily can be achieved, by using an Allure™ Biphenyl column under simple isocratic conditions. High retention capacity, similar to that of an ODS phase, also is demonstrated; a useful feature unavailable from conventional phenyl phases. By increasing π-π interactions, the Allure™ Biphenyl stationary phase offers a unique and more effective alternative to hydrophobic alkyl phases for resolving chemically similar unsaturated compounds, such as steroids.

References
1. *Superior Separations of Unsaturated Compounds by HPLC* Restek Advantage 2005v4 (lit. cat.# 580022).
2. *Improved HPLC Analysis of Steroids* Restek Application Note (lit. cat.# 580020).
3. Lake, R., and Wittrig, R., *Increasing HPLC Retention and Selectivity for Unsaturated Compounds, Using π-π Interactions* Pharmaceutical Canada, June 2006.

References 1&2 available on request.

Allure™ Biphenyl Columns

3µm Column, 2.1mm	cat. #
30mm	9166332
50mm	9166352
100mm	9166312
3µm Column, 3.2mm	
30mm	9166333
50mm	9166353
100mm	9166313
3µm Column, 4.6mm	
30mm	9166335
50mm	9166355
100mm	9166315

3µm Column, 2.1mm	
30mm (with Trident™ Inlet Fitting)	9166332-700
50mm (with Trident™ Inlet Fitting)	9166352-700
100mm (with Trident™ Inlet Fitting)	9166312-700
3µm Column, 3.2mm	
30mm (with Trident™ Inlet Fitting)	9166333-700
50mm (with Trident™ Inlet Fitting)	9166353-700
100mm (with Trident™ Inlet Fitting)	9166313-700
3µm Column, 4.6mm	
30mm (with Trident™ Inlet Fitting)	9166335-700
50mm (with Trident™ Inlet Fitting)	9166355-700
100mm (with Trident™ Inlet Fitting)	9166315-700
Allure™ Biphenyl Guard Cartridges	
10 x 2.1mm	916650212
10 x 4.0mm	916650210
20 x 2.1mm	916650222
20 x 4.0mm	916650220

New Rxi™-1ms GC Capillary Column

For Low Level GC/MS Analysis

By Robert Freeman, Environmental Innovations Chemist

- Inert, low-bleed column for reliable results from low-level GC/MS analyses.
- Save time – analyze acidic and basic compounds under the same conditions.
- Guaranteed reproducible performance, column to column.

The second column in our new Rxi™ GC column line – the Rxi™-1ms column – will provide the same outstanding performance as the Rxi™-5ms column, with equally superior inertness, ultra-low bleed, and excellent batch to batch reproducibility.

Our first test for this 100% dimethylpolysiloxane phase column was an analysis of a complex mixture of semivolatile organic compounds. The extensive target list was comprised of many classes of compounds including chloroacetanilides, chlorotriazines, triazinones, uracils, polcyclic aromatic hydrocarbons, and phthalates. Figure 1 shows peak shape and selectivity are equally good for all of these diverse compounds, and all are eluted in an acceptable analysis time.[1]

Excellent Inertness

In addition to analyzing these compounds, we analyzed an acidic compound (2,4-dinitrophenol) and a basic compound (pyridine), each at 0.5ng on column, to assess column inertness. Column activity reveals itself through poor response and peak tailing for such active compounds, and these two compounds present both varying difficulties in a GC/MS analysis and differing modes of degradation. Figure 2 shows the excellent peak shapes and responses for these compounds on the 30m x 0.25mm ID, 0.25µm film column.

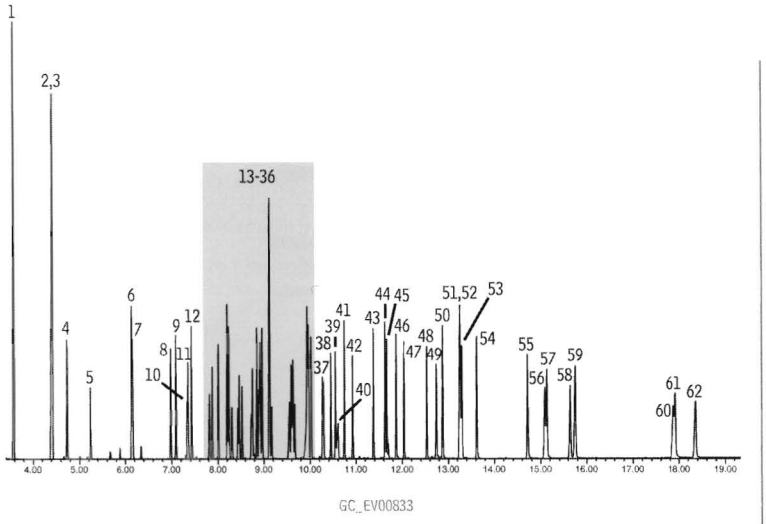
Phenols are notorious for breakdown and peak tailing, caused by interaction with the surface of an active inlet liner or an active column. Nitrophenols and pentachlorphenol, for example, very often exhibit poor peak shape and/or poor response. Figure 3 shows the 30m x 0.25mm ID, 0.25µm Rxi™-1ms column provides very good peak shapes for phenols. Peak responses are well above method requirements.

Ultra-Low Bleed

In addition to excellent inertness, Rxi™-1ms columns exhibit very low bleed. Figure 4 is focused on the end of the chromatogram for semivolatiles. At 330°C, bleed is much lower than the signals for 0.5ng of target analytes. This exceptional signal-to-noise differential for late eluting compounds assures better detection limits.

Figure 1 Excellent selectivity and peak shapes for common drinking water semivolatiles at 10ng, using an Rxi™-1ms column.

1. 2-fluorophenol (surr.)
2. bis(2-chloroethyl)ether
3. phenol-d6 (surr.)
4. 1,4-dichlorobenzene-d4 (int. std.) •
5. nitrobenzene-d5 (surr.)
6. naphthalene-d8 (int. std.) •
7. naphthalene
8. 1-methylnaphthalene
9. 2-methylnaphthalene
10. hexachlorocyclopentadiene
11. EPTC
12. 2-fluorobiphenyl (surr.)
13. 2,6-dinitrotoluene
14. dimethylphthalate
15. acenaphthylene
16. acenaphthene-d10 (int. std.) •
17. acenaphthene
18. 2,4-dinitrotoluene
19. 1-naphthalenamine
20. molinate
21. 2-naphthalenamine
22. 5-nitro-o-toluidine
23. diethylphthalate
24. fluorene
25. propachlor
26. diphenylamine
27. 2,4,6-tribromophenol (surr.)
28. simazine
29. prometon
30. atrazine
31. hexachlorobenzene
32. 4-aminobiphenyl
33. terbacil
34. phenanthrene-d10 (int. std.) •
35. phenanthrene
36. anthracene
37. metribuzin
38. acetochlor
39. alachlor
40. bromacil
41. di-*n*-butylphthalate
42. metolachlor
43. fluoranthene
44. pyrene
45. butachlor
46. *p*-terphenyl-d14 (surr.)
47. *p*-dimethylaminoazobenzene
48. benzyl butyl phthalate
49. 2-acetylaminofluorene
50. bis(2-ethylhexyl)adipate
51. benzo(a)anthracene
52. chrysene-d12 (int. std.) •
53. chrysene
54. bis(2-ethylhexyl)phthalate
55. di-*n*-octylphthalate
56. benzo(b)fluoranthene
57. benzo(k)fluoranthene
58. benzo(a)pyrene
59. perylene-d12 (int. std.) •
60. indeno(1,2,3-cd)pyrene
61. dibenzo(a,h)anthracene
62. benzo(ghi)perylene



Column:	Rxi™-1ms, 30m, 0.25mm ID, 0.25µm (cat.# 13323)
Sample:	US EPA Method 525.2 mix: custom 525.2 calibration mix, SV Internal Standard Mix (cat.# 31206), B/N Surrogate Mix (4/89 SOW) (cat.# 31024), Acid Surrogate Mix (4/89 SOW) (cat.# 31025)
Inj.:	1.0µL, 10µg/mL each analyte (internal standards 100µg/mL), split (10:1) 4mm Drilled Uniliner® inlet liner (hole at bottom) (cat.# 20756)
Instrument:	Agilent 6890
Inj. temp.:	250°C
Carrier gas:	helium, constant flow
Flow rate:	1.2mL/min.
Oven temp.:	50°C (hold 1 min.) to 265°C @ 20°C/min., to 330°C @ 6°Cmin. (hold 1 min.)
Det.:	Agilent 5973 MSD
Transfer line temp.:	280°C
Scan range:	35-550 amu
Solvent delay:	3.20 min.
Tune:	DFTPP
Ionization:	EI

• Internal standards at 100ng on-column.

Figure 2 An Rxi™-1ms column has excellent selectivity for basic or acidic compounds, under the same conditions. (0.5ng each; extracted ion chromatograms).

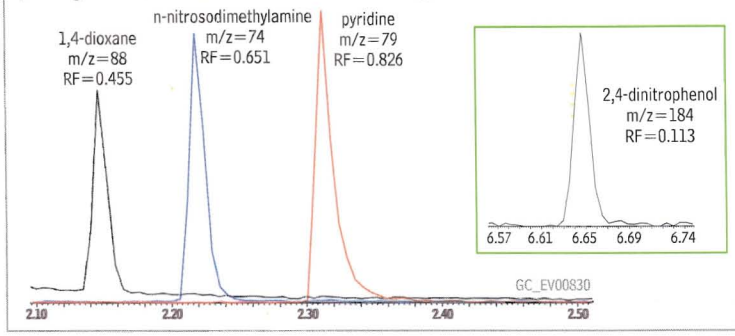
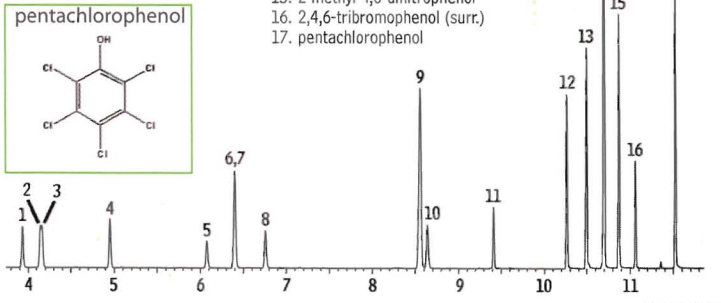


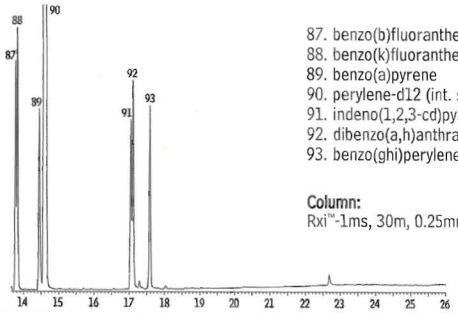
Figure 3 Acidic analytes at 5.0ng on an Rxi™-1ms column (extracted ion chromatogram).

1. phenol
2. 2-chlorophenol-d4 (surr.)
3. 2-chlorophenol
4. 2-methylphenol (*o*-cresol)
5. 2-nitrophenol
6. 2,4-dimethylphenol3,5,6-d3 (surr.)
7. 2,4-dimethylphenol
8. 2,4-dichlorophenol
9. 3-nitro-*o*-xylene (int. std.)
10. 4-chloro-3-methylphenol
11. 2,4,6-trichlorophenol
12. 2,4-dinitrophenol
13. 4-nitrophenol
14. 2,3,4,5-tetrachlorophenol (int. std.)
15. 2-methyl-4,6-dinitrophenol
16. 2,4,6-tribromophenol (surr.)
17. pentachlorophenol



Column:	Rxi™-1ms, 30m, 0.25mm ID, 0.25µm (cat.# 13323)
Sample:	US EPA Method 528 Mix: Phenols Fortification Mix, EPA 528 (cat.# 31695), Internal Standard Mix, EPA 528 (cat.# 31696), Surrogate Standard Mix, EPA 528 (cat.# 31697)
Inj.:	1.0µL, 5µg/mL each analyte (internal standards 25µg/mL), split (10:1) 4mm Drilled Uniliner® inlet liner (hole at bottom) (cat.# 20771)
Instrument:	Agilent 6890
Inj. temp.:	250°C
Carrier gas:	helium, constant flow
Flow rate:	1.2mL/min.
Oven temp.:	70°C (hold 0.5 min.) to 130°C @ 8°C/min., to 300°C @ 50°C/min. (hold 1 min.)
Det.:	Agilent 5973 MSD

Figure 4 Bleed profile at 330°C of Rxi™-1ms at end of 5ng on-column standard.



87. benzo(b)fluoranthene
88. benzo(k)fluoranthene
89. benzo(a)pyrene
90. perylene-d12 (int. std.)
91. indeno(1,2,3-cd)pyrene
92. dibenzo(a,h)anthracene
93. benzo(ghi)perylene

Column:
Rxi™-1ms, 30m, 0.25mm ID, 0.25µm (cat.# 13323)

Based on these results, we highly recommend the new Rxi™-1ms column for low-level analyses that require a 100% dimethylpolysiloxane phase.

Rxi™-1ms Columns (fused silica)

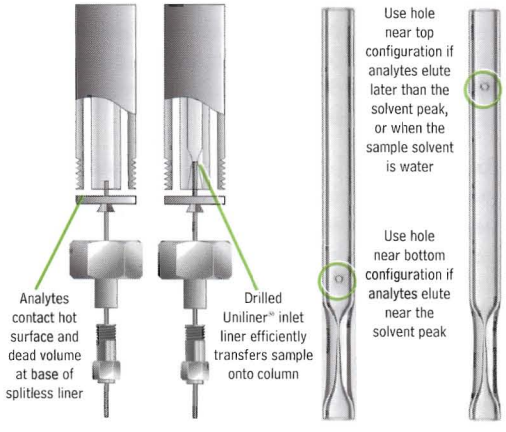
(Crossbond® 100% dimethylpolysiloxane)

ID	df (µm)	temp. limits	length	cat. #
0.18mm	0.18	-60 to 330/350°C	20-Meter	13302
0.20mm	0.33	-60 to 330/350°C	12-Meter	13397
0.20mm	0.33	-60 to 330/350°C	25-Meter	13398
0.20mm	0.33	-60 to 330/350°C	50-Meter	13399
0.25mm	0.25	-60 to 330/350°C	15-Meter	13320
0.25mm	0.25	-60 to 330/350°C	30-Meter	13323
0.25mm	0.25	-60 to 330/350°C	60-Meter	13326
0.25mm	0.50	-60 to 330/350°C	15-Meter	13335
0.25mm	0.50	-60 to 330/350°C	30-Meter	13338
0.25mm	0.50	-60 to 330/350°C	60-Meter	13341
0.25mm	1.00	-60 to 330/350°C	15-Meter	13350
0.25mm	1.00	-60 to 330/350°C	30-Meter	13353
0.25mm	1.00	-60 to 330/350°C	60-Meter	13356
0.32mm	0.25	-60 to 330/350°C	15-Meter	13321
0.32mm	0.25	-60 to 330/350°C	30-Meter	13324
0.32mm	0.25	-60 to 330/350°C	60-Meter	13327
0.32mm	0.50	-60 to 330/350°C	15-Meter	13336
0.32mm	0.50	-60 to 330/350°C	30-Meter	13339
0.32mm	0.50	-60 to 330/350°C	60-Meter	13342
0.32mm	1.00	-60 to 330/350°C	15-Meter	13351
0.32mm	1.00	-60 to 330/350°C	30-Meter	13354
0.32mm	1.00	-60 to 330/350°C	60-Meter	13357
0.53mm	0.50	-60 to 330/350°C	15-Meter	13337
0.53mm	0.50	-60 to 330/350°C	30-Meter	13340
0.53mm	1.00	-60 to 330/350°C	15-Meter	13352
0.53mm	1.00	-60 to 330/350°C	30-Meter	13355
0.53mm	1.50	-60 to 330/350°C	15-Meter	13367
0.53mm	1.50	-60 to 330/350°C	30-Meter	13370
0.53mm	1.50	-60 to 330/350°C	60-Meter	13373

restek innovation!

[1]The Drilled Uniliner®

To reduce the effects of surface activity in the injection port liner, and focus on the effects of the column on active analytes, we used a Drilled Uniliner® inlet liner for this work. This liner connects directly to the column, eliminating contact between the active compounds and active metal surfaces in the injector, and ensuring an inactive sample pathway for analyte transfer from the injection port to the column.



Analysis of Semivolatile Organics

Using the new Rxi™-5ms Capillary GC Column

by Robert Freeman, Environmental Innovations Chemist, and Christopher M. English, Innovations Group Leader

- Low column bleed, outstanding inertness, excellent column-to-column reproducibility.
- Symmetric peaks and good response factors for acidic or basic analytes.
- Resolve 93 analytes in less than 18 minutes.

Sub-nanogram Analysis of Semivolatile Organics

Analyzing basic or acidic semivolatile environmental pollutants at low nanogram-on-column concentrations puts demands on the entire analytical system. Using our new Rxi™-5ms column, we have developed an analytical procedure that assures good performance for both acids and bases.

Rxi™-5ms Column Offers Sensitivity for Acids and Bases

One of the most active basic compounds listed in semivolatiles methods is pyridine. This early-eluting compound can elicit poor performance in the injection port and on the column, and many currently available columns give a poor peak shape for pyridine. Columns with a slightly basic surface can perform well with pyridine, but will perform poorly with the acidic compounds, such as 2,4-dinitrophenol.

Figure 1 combines extracted ion chromatograms for the initial three US EPA Method 8270D target compounds, at 0.5ng per compound on-column. The extracted ion for 1,4-dioxane shows that injection port and oven conditions were optimized. The pyridine and N-nitrosodimethylamine peaks are symmetric, even at this low level of detection. An excessively tailing pyridine peak, or a pyridine peak smaller than that for 1,4-dioxane at the same concentration, would indicate on-column activity. With an Rxi™-5ms column, and the conditions listed for Figure 3, pyridine can be detected reliably at low concentrations.

Analytically, 2,4-dintrophenol is considered the most problematic compound in the Method 8270 target list. 2,4-Dinitrophenol and the other system performance check compounds (SPCC) - N-nitroso-di-*n*-propylamine, hexachlorocyclopentadiene, and 4-nitrophenol - must exhibit a minimum average response factor (RF) of 0.050. An optimized system generally will provide response factors greater than 0.1 for these compounds, but the lower the calibration curve for these compounds, the more difficult it is to achieve passing response factors. If any of these compounds fails to meet the Method 8270 response factor criterion, system maintenance must be performed to bring response factors to passing before samples can be

Figure 1 An Rxi™-5ms column provides sharp, easily quantified peaks for active analytes (extracted ion chromatograms).

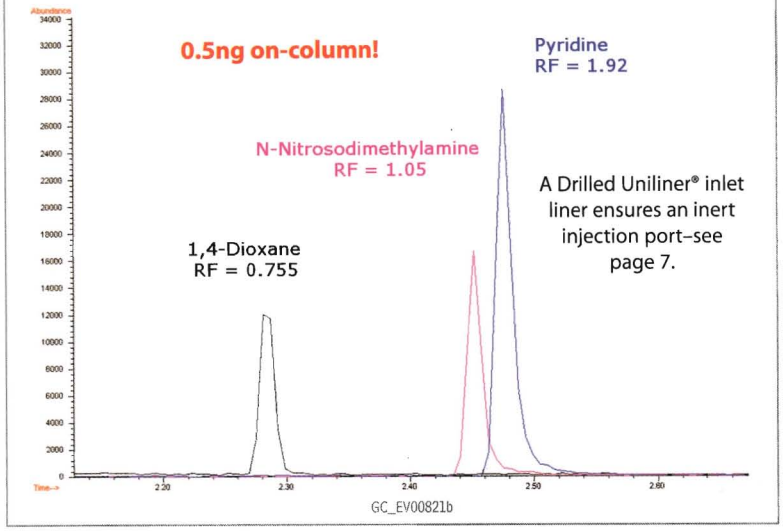


Figure 2 Excellent response for 0.5ng 2,4-dintrophenol on an Rxi™-5ms column.

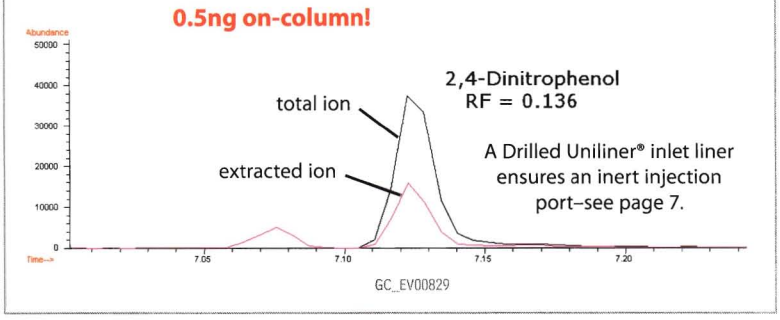
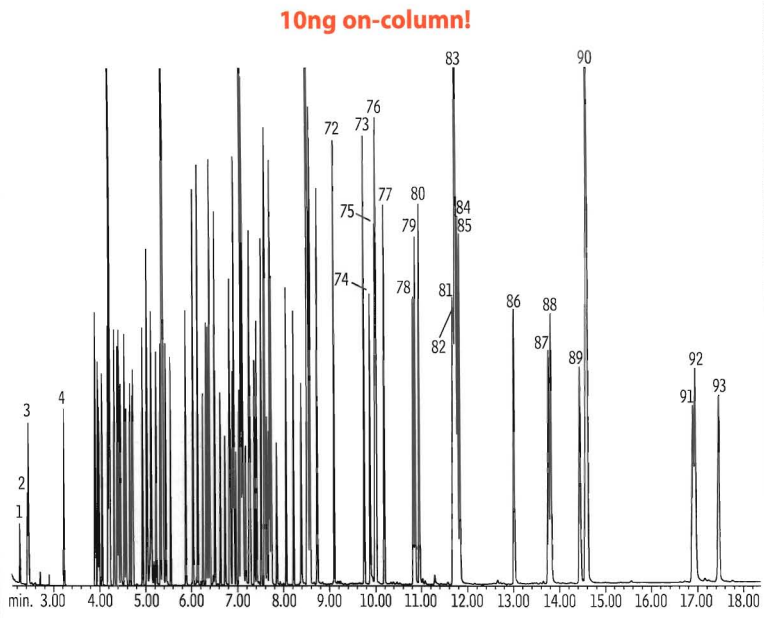


Figure 3 Separate 93 semivolatile compounds in less than 18 minutes, using an Rxi™-5ms column.



GC_EV00823

For complete identifications, please visit www.restek.com/rxi

1. 1,4-dioxane
2. N-nitrosodimethylamine
3. pyridine
4. 2-fluorophenol
5. phenol-d6
6. phenol
7. aniline
8. bis(2-chloroethyl) ether
9. 2-chlorophenol
10. 1,3-dichlorobenzene
11. 1,4-dichlorobenzene-d4
12. 1,4-dichlorobenzene
13. benzyl alcohol
14. 1,2-dichlorobenzene
15. 2-methylphenol
16. bis(2-chloroisopropyl) ether
17a. 4-methylphenol
17b. 3-methylphenol
18. N-nitroso-di-*n*-propylamine
19. hexachloroethane
20. nitrobenzene-d5
21. nitrobenzene
22. isophorone
23. 2-nitrophenol
24. 2,4-dimethylphenol
25. benzoic acid
26. bis(2-chloroethoxy)methane
27. 2,4-dichlorophenol
28. 1,2,4-trichlorobenzene
29. naphthalene-d8
30. naphthalene
31. 4-chloroaniline
32. hexachlorobutadiene
33. 4-chloro-3-methylphenol
34. 2-methylnaphthalene
35. 1-methylnaphthalene
36. hexachlorocyclopentadiene
37. 2,4,6-trichlorophenol
38. 2,4,5-trichlorophenol
39. 2-fluorobiphenyl
40. 2-chloronaphthalene
41. 2-nitroaniline
42. 1,4-dinitrobenzene
43. dimethyl phthalate
44. 1,3-dinitrobenzene
45. 2,6-dinitrotoluene
46. acenaphthylene
47. 1,2-dinitrobenzene
48. 3-nitroaniline
49. acenaphthene-d10
50. acenaphthene
51. 2,4-dinitrophenol
52. 4-nitrophenol
53. dibenzofuran
54. 2,4-dinitrotoluene
55. 2,3,4,6-tetrachlorophenol
56. 2,3,5,6-tetrachlorophenol
57. diethyl phthalate
58. 4-chlorophenyl phenyl ether
59. fluorene
60. 4-nitroanaline
61. 4,6-dinitro-2-methylphenol
62. N-nitrosodiphenylamine (as diphenylamine)
63. 1,2-diphenylhydrazine (as azobenzene)
64. 2,4,6-tribromophenol
65. 4-bromophenyl phenyl ether
66. hexachlorobenzene
67. pentachlorophenol
68. phenanthrene-d10
69. phenanthrene
70. anthracene
71. carbazole
72. di-*n*-butyl phthalate
73. fluoranthene
74. benzidine
75. pyrene-d10
76. pyrene
77. *p*-terphenyl-d14
78. 3,3'-dimethylbenzidine
79. butyl benzyl phthalate
80. bis(2-ethylhexyl) adipate
81. 3,3'-dichlorobenzidine
82. benzo(a)anthracene
83. chrysene-d12
84. chrysene
85. bis(2-ethylhexyl) phthalate
86. di-*n*-octyl phthalate
87. benzo(b)fluoranthene
88. benzo(k)fluoranthene
89. benzo(a)pyrene
90. perylene-d12
91. indeno(1,2,3-cd)pyrene
92. dibenzo(a,h)anthracene
93. benzo(ghi)perylene

Column:	Rxi™-5ms, 30m, 0.25mm ID, 0.25µm (cat.# 13423)
Sample:	US EPA Method 8270D mix: 8270 MegaMix™ (cat.# 31850), Benzoic Acid Standard (cat.# 31879), Benzidine Standard (cat.# 31852), Acid Surrogate Mix (cat.# 31025), B/N Surrogate Standard Mix (cat.# 31887), 1,4-Dioxane (cat.# 31853)
Inj.:	1.0µL, 10ppm each analyte (10ng on column), splitless (hold 0.1 min.) **4mm Drilled Uniliner® inlet liner (hole at bottom) (cat.# 20756)**
Instrument:	Agilent 6890
Inj. temp.:	250°C
Carrier gas:	helium, constant flow
Flow rate:	1.2mL/min.
Oven temp.:	50°C (hold 0.5 min.) to 265°C @ 25°C/min., to 330°C @ 6°C/min. (hold 2 min.)
Det.:	Agilent 5973 GC/MS
Transfer line temp.:	280°C
Scan range:	35-550 amu
Solvent delay:	2 min.
Tune:	DFTPP
Ionization:	EI

analyzed. Figure 2 shows the inertness of the Rxi™-5ms column, which exhibits a response factor of 0.136 for 0.5ng on-column of 2,4-dinitrophenol.

The total ion chromatogram for our optimized analysis is shown in Figure 3. There are at least five scans across each target analyte, which assures good spectral integrity and good peak shape, and the last compound is eluted in less than 18 minutes.

The Result

The Rxi™-5ms column introduces a new generation of Restek columns that exhibit low bleed, outstanding inertness, and excellent column-to-column reproducibility.

An Rxi™-5ms column, used in an optimized system, provides excellent chromatography for Method 8270 semivolatile compounds, including difficult-to-analyze acidic or basic compounds, at low on-column concentrations. These new columns give the performance needed, at the sensitivity required, column after column.

Rxi™-5ms Columns (fused silica)

(Crossbond® 5% diphenyl / 95% dimethyl polysiloxane)

ID	df (µm)	temp. limits	length	cat. #
0.18mm	0.18	-60 to 330/350°C	20-Meter	13402
0.18mm	0.36	-60 to 330/350°C	20-Meter	13411
0.20mm	0.33	-60 to 330/350°C	12-Meter	13497
0.20mm	0.33	-60 to 330/350°C	25-Meter	13498
0.20mm	0.33	-60 to 330/350°C	50-Meter	13499
0.25mm	0.25	-60 to 330/350°C	15-Meter	13420
0.25mm	0.25	-60 to 330/350°C	30-Meter	13423
0.25mm	0.25	-60 to 330/350°C	60-Meter	13426
0.25mm	0.50	-60 to 330/350°C	15-Meter	13435
0.25mm	0.50	-60 to 330/350°C	30-Meter	13438
0.25mm	0.50	-60 to 330/350°C	60-Meter	13441
0.25mm	1.00	-60 to 330/350°C	15-Meter	13450
0.25mm	1.00	-60 to 330/350°C	30-Meter	13453
0.25mm	1.00	-60 to 330/350°C	60-Meter	13456
0.32mm	0.25	-60 to 330/350°C	15-Meter	13421
0.32mm	0.25	-60 to 330/350°C	30-Meter	13424
0.32mm	0.25	-60 to 330/350°C	60-Meter	13427
0.32mm	0.50	-60 to 330/350°C	15-Meter	13436
0.32mm	0.50	-60 to 330/350°C	30-Meter	13439
0.32mm	1.00	-60 to 330/350°C	15-Meter	13451
0.32mm	1.00	-60 to 330/350°C	30-Meter	13454
0.32mm	1.00	-60 to 330/350°C	60-Meter	13457
0.53mm	0.25	-60 to 330/350°C	15-Meter	13422
0.53mm	0.25	-60 to 330/350°C	30-Meter	13425
0.53mm	0.50	-60 to 330/350°C	15-Meter	13437
0.53mm	0.50	-60 to 330/350°C	30-Meter	13440
0.53mm	1.00	-60 to 330/350°C	15-Meter	13452
0.53mm	1.00	-60 to 330/350°C	30-Meter	13455
0.53mm	1.50	-60 to 330/350°C	15-Meter	13467
0.53mm	1.50	-60 to 330/350°C	30-Meter	13470

tech **tip**

A Drilled Uniliner® inlet liner helps ensure reliable results for active compounds—see information on page 7.

8-Minute GC Analysis of Residual Solvents

Using an Rtx®-624 (G43) / Rtx®-WAX (G16) Column Pair

By Rick Lake, Pharmaceutical Innovations Chemist

- Dual-column detection/confirmation in 8 minutes.
- Columns produce desired selectivity and stable retention.
- Excellent peak shape and sensitivity, for reliable information.

The International Conference on Harmonization (ICH) publishes a guideline (Q3C) listing amounts of solvent residues that are acceptable in drug products and drug substances. The complete ICH list of regulated solvents, 61 compounds of differing chemical properties, is a challenge for separation on any single GC phase, as critical coelutions exist. Typically, residual solvents are identified by assaying samples and matching retention times with reference standards. If a response greater than the regulatory limit is obtained in a retention time window, a second sample is analyzed to confirm the compound's identity, using a column that has alternate selectivity. In some cases, GC/MS is employed for analyte verification. Assays for verification can be laborious and time intensive, and add unnecessary cost.

In the ICH guideline, residual solvents are grouped according to their toxicity. Class 1 compounds are carcinogens that pose a risk to both consumers and the environment. The use of these solvents is to be avoided but, if they are used, their use must be tightly controlled to ensure only trace level impurities in the final product. Class 2 compounds are non-genotoxic animal carcinogens, and concentrations of these compounds should be limited in pharmaceutical actives and products. Class 3 compounds have low toxic potential, and concentrations up to 0.5% are acceptable. Therefore, Class 3 compounds can be assayed by non-specific techniques, such as weight loss on drying. Because Class 2 compounds are the most likely prospects for GC analysis, we selected Residual Standards Class 2 Mix A and Residual Standards Class 2 Mix B (cat.#s 36271 and 36272, respectively) as the analytes for this work.

Because of advances in headspace technology – mainly dynamic sampling techniques – greater sensitivity now is achievable with this approach[1], and this makes a comprehensive dual-column assay feasible. By simultaneously using two columns with differing selectivities, e.g., a G43 column (Rtx®-1301 or Rtx®-624) and a G16 column (Rtx®-Wax or Stabilwax®), a single injection can be used both to detect residual solvents and to confirm their identities. Even with two columns, however, the complexity of the sample list makes it impossible for a single temperature program to provide the flexibility needed to resolve all compounds on each column. To overcome this barrier, we used a Tekmar HT3 dynamic headspace sampler and an Agilent 6890 GC equipped with a Gerstel Modular Accelerated Column Heater (MACH) System. One of the latest advances in fast GC technology, the MACH System incorporates columns encased individually in thermally controlled bundles and heated externally from the main GC oven (Figure 1).[2] This independent, low thermal mass configuration allows independent, very rapid temperature ramps, upward or downward.

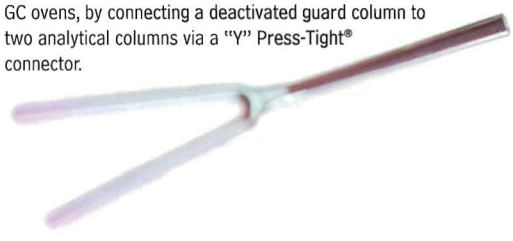
Collected analytes were directed to the injection port, then were split onto the two columns via a "Y" Press-Tight® connector. Independent temperature programs for each column separated the analytes for detection on dual FIDs. Using our two columns in this novel and simple-to-use setup, we resolved all compounds in the combined reference mixes in less than 8 minutes (Figure 2) – a result not possible with a conventional GC system. There was one critical co-elution on each column, but these did not involve the same compounds, and thus posed no practical problem. Also, with the low thermal mass of the

Figure 1 Two column modules in a Gerstel MACH column heating system.

The MACH system allows independent temperature programming of up to four columns, simultaneously.

tech tip

Dual column assays also can be performed in conventional GC ovens, by connecting a deactivated guard column to two analytical columns via a "Y" Press-Tight® connector.



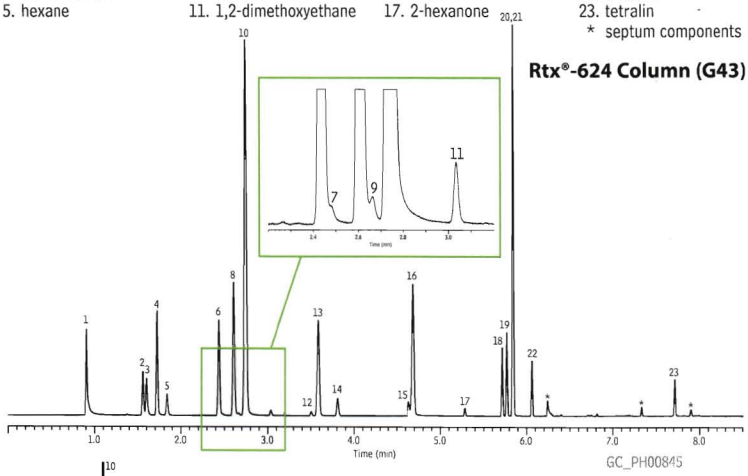
Universal "Y" Press-Tight® Connectors

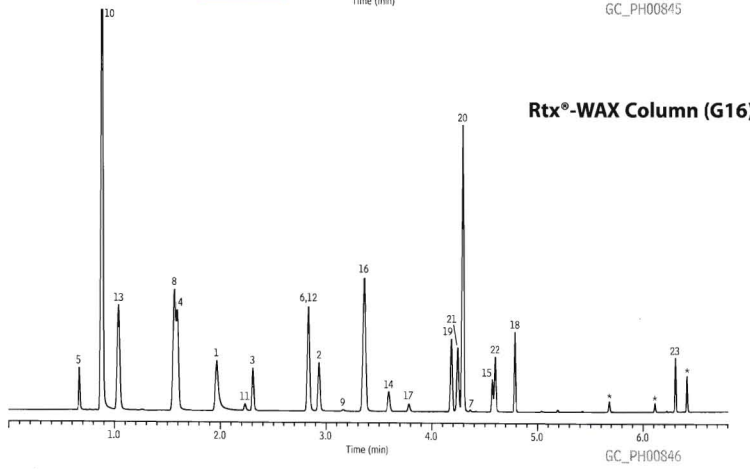
Description	cat. #
Universal "Y" Press-Tight® Connector	20405

Figure 2 Detect and confirm residual solvents with one injection, using Restek columns in a Gerstel MACH System.

1. methanol
2. acetonitrile
3. dichloromethane
4. *trans*-1,2-dichloroethylene
5. hexane
6. *cis*-1,2-dichloroethylene
7. nitromethane
8. tetrahydrofuran
9. chloroform
10. cyclohexane
11. 1,2-dimethoxyethane
12. trichloroethylene
13. methylcyclohexane
14. 1,4-dioxane
15. pyridine
16. toluene
17. 2-hexanone
18. chlorobenzene
19. ethyl benzene
20. *m*-xylene
21. *p*-xylene
22. *o*-xylene
23. tetralin

* septum components





Headspace Conditions - see our web page: www.restek.com/buzz

Instrument Conditions

Instrument:	Agilent 6890 with Gerstel Modular Accelerated Column Heater (MACH)
Column:	Column 1: Rtx®-624 (G43) 20m, 0.18mm ID, 1.00µm (cat.# 40924) Column 2: Rtx®-WAX (G16) 20m, 0.18mm ID, 0.4µm (custom)
Sample:	cat.# 36271 and cat.# 36272 diluted in 0.5mL DI water/~0.2g sodium sulfate in 20mL headspace vial; components at 0.10 - 6.00µg - see our web page.
Inj.:	headspace trap injection, split ratio 20:1, flow split after injector to two columns, using a 0.32mm ID intermediate polarity deactivated guard column (cat.# 10044) and a "Y" Press-Tight® connector (cat.# 20405)
Inj. temp.:	220°C
Carrier:	helium, constant flow
Flow rate:	Column 1: 0.85mL/min.; Column 2: 0.99mL/min.
Oven temp.:	250°C
Gerstel temps.:	Column 1: 50°C (2 min.) to 80°C at 20°C/min. (1 min.), to 200°C at 40°C/min. (2 min.) Column 2: 35°C (2 min.) to 60°C at 100°C/min. (1 min.), to 200°C at 40°C/min. (2 min.)
Det.:	FID at 250°C, hydrogen: 40mL/min., air: 450mL/min., makeup gas: 45mL/min.

1. Restek Advantage 2006 vol. 1, pp.14-15 (2006); request: lit. cat.# 580035.
2. Direct inquiries about the Gerstel MACH System to Gerstel Inc. Phone: 410-247 5885; e-mail: sales@gerstelus.com

MACH System modules, the cooldown and equilibration time between samples is considerably shorter than with a conventional GC oven.

Dynamic headspace sampling coupled with a Gerstel MACH column heating system makes possible rapid, comprehensive assays of residual solvents. By using other column combinations and other independent temperature programs, this system can be adapted to quickly resolve other complex mixes.

Residual Solvents Class 2 - Mix A (15 components)
In dimethyl sulfoxide, 1mL/ampul
cat. # 36271 (ea.)

Residual Solvents Class 2 - Mix B (8 components)
In dimethyl sulfoxide, 1mL/ampul
cat. # 36272 (ea.)

Residual Solvents Class 2 - Mix C (8 components)
In dimethyl sulfoxide, 1mL/ampul
cat. # 36273 (ea.)

European Pharmacopoeia/ICH Q3C(M) Class 2 Mix C (14 components)
In dimethyl sulfoxide, 1mL/ampul
cat. # 36274 (ea.)

European Pharmacopoeia/ICH Q3C(M) Class 2 Mix A (6 components)
In dimethyl sulfoxide, 1mL/ampul
cat. # 36275 (ea.)

Fused Silica Guard Columns/Transfer Lines

Nominal ID	Nominal OD	length	cat. #
0.32mm	0.45 ± 0.04mm	5-Meter	10044
0.32mm	0.45 ± 0.04mm	5-Meter	10044-600

gc column **ordering** info

To order Gerstel MACH GC Columns, call:
800-413-8160
410-247-5885
e-mail: sales@gerstelus.com

Rtx®-624 (G43) (fused silica)
(Crossbond® 6% cyanopropylphenyl/94% dimethyl polysiloxane)

ID	df (µm)	temp. limits	length	Module	Gerstel cat. #
0.18mm	1.00	-20 to 240°C	20-Meter	5"	015200-019-GI
0.18mm	1.00	-20 to 240°C	20-Meter	3"	015200-020-GI

Rtx®-WAX (G16) (fused silica)

ID	df (µm)	temp. limits	length	Module	Gerstel cat. #
0.18mm	0.4	-20 to 250°C	20-Meter	5"	015200-021-GI
0.18mm	0.4	-20 to 250°C	20-Meter	3"	015200-022-GI

Simple, Optimized HPLC Analysis of Catecholamines

Increase Retention by Using an Allure™ PFP Propyl Column

By Rick Lake, Pharmaceutical Innovations Chemist, and Bruce Albright, HPLC Innovations Chemist

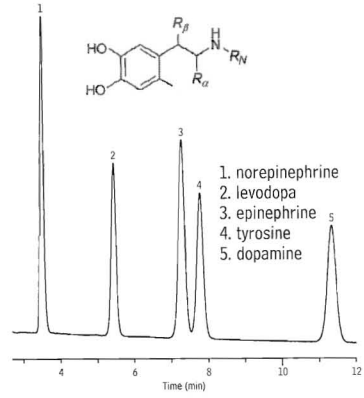
- No derivatization or ion-pairing—save time, ensure reproducible results.
- Excellent retention and resolution of low molecular weight amine compounds.
- Excellent peak shapes for reliable quantification of basic compounds.

Biogenic amines are low molecular weight intercellular messengers that relay much of the body's chemical signaling. Many synthesized drug compounds are chemically similar to these very biologically active compounds, including stimulants, hallucinogens, antidepressants, and bronchodilators.

One group of biogenic amines, the catecholamines (Figure 1), traditionally have been assayed by GC or HPLC, but either approach requires modifications. Derivatization is necessary for GC analysis, and stability issues can pose a problem. Limited retention on hydrophobic alkyl (ODS) or polar embedded (cyano) HPLC phases makes derivatization or ion-pairing techniques necessary. These modified HPLC techniques are laborious and disrupt reproducibility, and many derivatizing reagents are not LC/MS compatible.

Pentafluorophenyl HPLC phases show greater retention for compounds that have electrophilic properties, like protonated amine groups in basic compounds, and a propyl spacer between the functional group and the silica surface - a pentafluorophenyl propyl phase - further increases retention. Consequently, when an acidic mobile phase is used to induce protonation of the analytes' amine groups, the Allure™ PFP Propyl phase makes possible a simple reversed phase HPLC analysis (Figure 1). A nearly 100% aqueous mobile phase is needed, but retention of norepinephrine, the first eluting analyte, is sufficient. By changing the organic modifier, differing selectivities can be achieved (Figure 2), giving the analyst more flexibility in optimizing specific separations. By using an Allure™ PFP Propyl column, an analyst can achieve simple, reproducible analyses of catecholamines or similar low molecular weight polar compounds.

Figure 1 Superior retention of catecholamines on an Allure™ PFP Propyl column—better separations, without ion pairing.



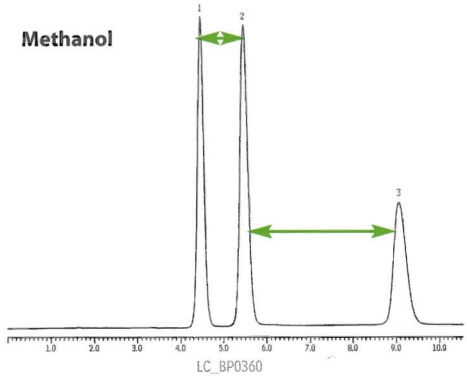
Sample:
Inj.: 10µL
Conc.: 40µg/mL each component
Sample diluent: mobile phase
Sample temp.: ambient

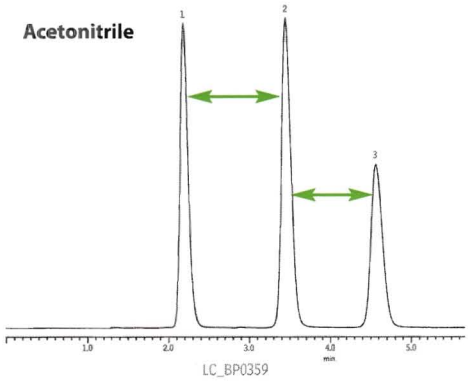
Column: Allure™ PFP Propyl (cat.# 9169565)
Dimensions: 150 x 4.6mm
Particle size: 5µm
Pore size: 60Å

Conditions:
Mobile phase: 50mM ammonium phosphate (pH 3.0, adjusted with formic acid)
Flow: 1mL/min.
Temp.: 35°C
Det.: UV @ 266nm

LC_BA0349

Figure 2 Changing organic modifiers alters selectivity for catecholamines, for more flexibility in optimizing separations.





Peak List
1. levodopa
2. epinephrine
3. dopamine

Sample:
Inj.: 10µL
Conc.: 100µg/mL each component
Sample diluent: mobile phase

Column: Allure™ PFP Propyl (cat.# 9169565)
Dimensions: 150 x 4.6 mm
Particle size: 5µm
Pore size: 60Å

Conditions:
Mobile phase: 20mM ammonium formate (pH 3): methanol, 90:10
or
20mM ammonium formate (pH 3): acetonitrile, 90:10
Flow: 1mL/min.
Temp.: ambient
Det.: UV @ 280 nm

Allure™ PFP Propyl, 5µm Columns

5µm Column, 4.6mm	cat. #
150mm	9169565
150mm (with Trident™ Inlet Fitting)	9169565-700

Allure™ PFP Propyl Guard Cartridges

Allure™ PFP Propyl	qty.	cat. #
10 x 2.1mm	3-pk.	916950212
10 x 4.0mm	3-pk.	916950210
20 x 2.1mm	2-pk.	916950222
20 x 4.0mm	2-pk.	916950220

80% Faster GC/MS Analysis of Essential Oils

Using a 10m x 0.10mm ID Rtx®-5 Column

by Novalina Lingga, Ph.D., Application Chemist, Shimadzu Asia Pacific, Singapore and Eberhardt Kuhn, Ph.D., International Marketing Specialist, Restek Corp.

- 5x greater sample throughput.
- Sharply reduced cost per analysis.
- Resolution and elution orders are not changed.

Essential oils are key components of perfumes, soaps, and other cosmetic products, and they find extensive use in aromatherapy. Because they have high market value, essential oils are subject to adulteration with less expensive impurities. It is, therefore, important to have reliable analytical methods to determine the purity of essential oils.

Typical analysis times for these complex samples, using a 30m x 0.25mm x 0.25µm df Rtx®-5SilMS column and "conventional" GC/MS conditions, are 18 minutes for bergamot oil and 30 minutes for patchouli oil (the analyses are posted on our website). Figure 1 shows these analyses optimized for speed, using a 10m x 0.10mm x 0.10µm df Rtx®-5 column. Analysis times were reduced to approximately 3.5 minutes for bergamot oil and 5.5 minutes for patchouli oil. This 80% reduction in analysis time increases sample throughput by a factor of 5, without sacrificing resolution or accuracy!

Relative to conventional analyses, resolution in the fast analyses is essentially unchanged for bergamot oil, and actually is slightly better for patchouli oil. Because the phase ratio (β) was kept constant at 250, the elution order of the oil components is identical for both fast and conventional analyses, allowing easy peak identification and comparison.

These results demonstrate the potential for greatly increased throughput for essential oils by using a shorter, smaller diameter column. The cost savings make this a desirable improved method for any laboratory.

Rtx®-5 Column (fused silica)

(5% diphenyl/95% dimethyl polysiloxane)

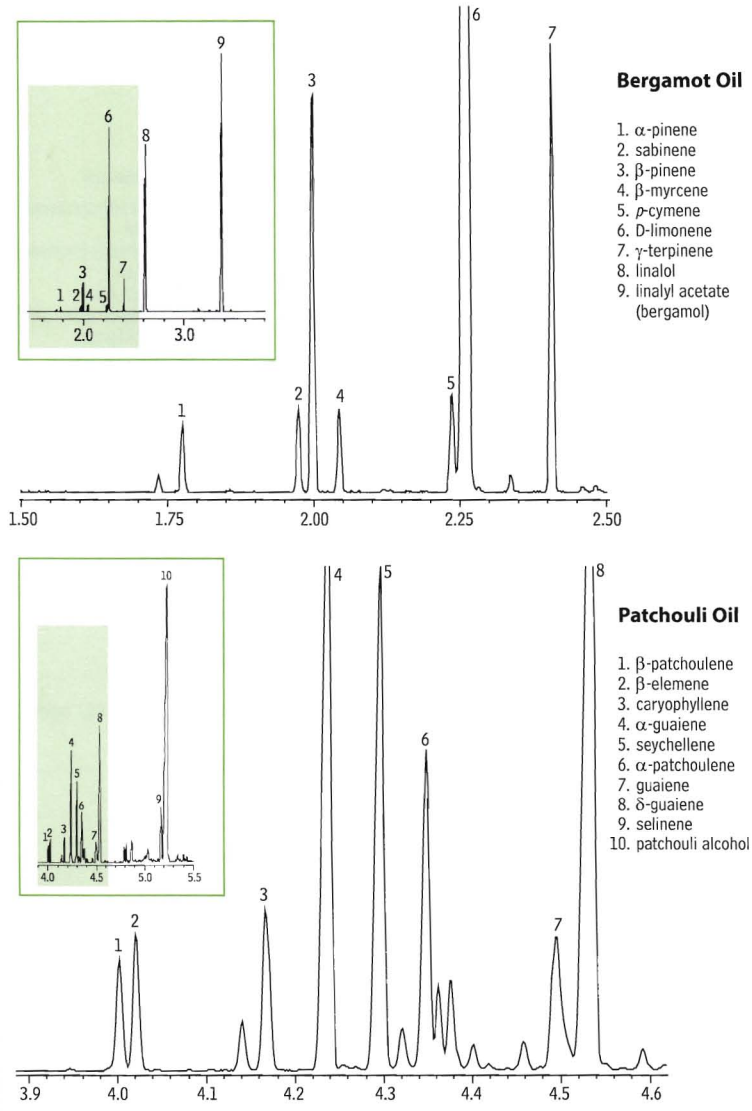
ID	df (µm)	temp. limits	length	cat. #
0.10mm	0.10	-60 to 330/350°C	10-Meter	41201

Rtx®-5Sil MS Column (fused silica)

(Selectivity similar to 5% diphenyl/95% dimethyl polysiloxane)

ID	df (µm)	temp. limits	length	cat. #
0.25mm	0.25	-60 to 330/350°C	30-Meter	12723

Figure 1 Fast GC/MS analysis of essential oils: equivalent to conventional chromatography, 80% faster.



Column:	Rtx®-5, 10m x 0.10mm ID, 0.10µm (cat. # 41201)
Sample:	bergamot oil, 5% in acetone, 0.2µL injection
Instrument:	Shimadzu GCMS-QP2010
Injector:	split/splitless, 280°C
Injection mode:	split, split ratio = 800:1
Carrier gas:	helium
Flow control mode:	constant linear velocity (60.0 cm/sec)
Column inlet pressure:	421.5kPa @ 40°C
Column oven:	40°C – 30°C/min. – 300°C
Interface:	direct, 280°C
Ionization:	EI
Mass range:	40-500 amu

Chromatograms courtesy of Dr. Novalina Lingga, Shimadzu Asia Pacific, Singapore

Analytical Reference Materials for SOM01.1

by Ken Herwehe, Analytical Reference Materials Product Manager

- SIM compounds included in reformulated OLC 03.2 SVOA Deuterated Monitoring Compounds (DMC) mix for semivolatiles (cat.# 31810).
- SOM01.1 SVOA MegaMix™ (cat.# 33005) combines 67 semivolatiles in a single mix.
- Both ketones and non-ketones included in reformulated SOM01.1 VOA DMC Kit (cat.# 30630).

SOM01.1 defines methods for isolating, detecting, and quantitatively measuring 52 trace and low/medium level volatile, 67 semivolatile, 21 pesticide, and 9 Aroclor® target compounds in water or soil/sediment environmental samples. This document incorporates major changes to the organic methods, including separating the pesticide and Aroclor® methods, reformulating the deuterated monitoring compounds for volatiles and semivolatiles, and including selected ion monitoring (SIM) analysis. SOM01.1 calls for gas chromatography/mass spectrometry (GC/MS) and gas chromatography/electron capture detector (GC/ECD) methods for analyzing the target compounds.

please note

1. Ketone and non-ketone mixes can be purchased individually.
2. Ketone and non-ketone mixes are prepared at 500µg/mL.
3. Our methanol-d solvent (CH_3-OD; MW33)does not interfere with MS scans. Per-deuterated methanol (CD_3-OD; MW 36) interferes with MS scans.

did you know?

Our new Rxi™ capillary columns are ideal for analyses of environmental samples. For example applications, refer to the articles on **pages 6-7 and 8-9**.

For additional information, visit our website at **www.restek.com/rxi**

Volatiles: Calibration Mixes

OLC 03.2 VOA MegaMix™ (42 components)

benzene
bromochloromethane
bromodichloromethane
bromoform
carbon disulfide
carbon tetrachloride
chlorobenzene
chloroform
cyclohexane
dibromochloromethane (chlorodibromomethane)
1,2-dibromo-3-chloropropane
1,2-dibromoethane (EDB)
1,2-dichlorobenzene
1,3-dichlorobenzene
1,4-dichlorobenzene
1,1-dichloroethane
1,2-dichloroethane
1,1-dichloroethylene
cis-1,2-dichloroethylene
trans-1,2-dichloroethylene
1,2-dichloropropane
cis-1,3-dichloropropene
trans-1,3-dichloropropene
ethylbenzene
isopropylbenzene (cumene)
methyl acetate
methylcyclohexane
methyl *tert*-butyl ether (MTBE)
methylene chloride (dichloromethane)
styrene
1,1,2,2-tetrachloroethane
tetrachloroethylene
toluene
1,2,3-trichlorobenzene
1,2,4-trichlorobenzene
1,1,1-trichloroethane
1,1,2-trichloroethane
trichloroethylene
1,1,2-trichlorotrifluoroethane (Freon® 113)
m-xylene*
o-xylene
p-xylene*

2,000µg/mL each (**m*- & *p*-xylene at 1,000µg/mL) in P&T methanol, 1mL/ampul
cat. # 30492 (ea.)

1,4-Dioxane

2,000µg/mL in P&T methanol, 1mL/ampul
cat. # 30287 (ea.)

502.2 Calibration Mix #1 (gases)

bromomethane
chloroethane
chloromethane
dichlorodifluoromethane
trichlorofluoromethane
vinyl chloride

200µg/mL each in P&T methanol, 1mL/ampul
cat. # 30439 (ea.)

2,000µg/mL each in P&T methanol, 1mL/ampul
cat. # 30042 (ea.)

VOA Calibration Mix #1 (ketones)

acetone
2-butanone
2-hexanone
4-methyl-2-pentanone

5,000µg/mL each in P&T methanol:water (90:10), 1mL/ampul
cat. # 30006 (ea.)

Volatiles: DMC

SOM01.1 VOA Non-Ketone Deuterated Monitoring Compounds (11 components)

benzene-d6
chloroethane-d5
chloroform-d
1,2-dichlorobenzene-d4
1,2-dichloroethane-d4
1,1-dichloroethene-d2
1,2-dichloropropane-d6
1,3-dichloropropene-d4*
1,1,2,2-tetrachloroethane-d2
toluene-d8
vinyl chloride-d3

500µg/mL each in deuterated methanol (MeOD), 1mL/ampul
cat. # 30624 (ea.)

*Mix of *cis* and *trans* isomers. Exact proportions will be reported on the data sheet.

SOM01.1 VOA Ketone Deuterated Monitoring Compounds

2-butanone-d5
2-hexanone-d5

500µg/mL each in deuterium oxide (D_2O), 1mL/ampul
cat. # 30625 (ea.)

SOM01.1 VOA DMC Kit

kit

30624: Non-Ketones 500µg/mL
30625: Ketones 500µg/mL

1mL each of these mixtures.
cat. # 30630 (kit)

1,4-Dioxane-d8

2,000µg/mL in P&T methanol, 1mL/ampul
cat. # 30614 (ea.)

Semivolatiles: DMC

OLC 03.2 SVOA Deuterated Monitoring Compounds (DMC) (16 components)

acenaphthylene-d8
anthracene-d10
benzo(a)pyrene-d12
4-chloroaniline-d4
bis-(2-chloroethyl)ether-d8
2-chlorophenol-d4
2,4-dichlorophenol-d3
dimethylphthalate-d6
4,6-dinitro-methylphenol-d2
fluorene-d10
4-methylphenol-d8
nitrobenzene-d5
2-nitrophenol-d4
4-nitrophenol-d4
phenol-d5
pyrene-d10

2,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 31810 (ea.)

SOM01.1 Deuterated Monitoring Compound Mix SIM Compounds

fluoranthene-d10
2-methylnaphthalene-d10

2,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 33913 (ea.)

SOM01.1 Deuterated Monitoring Compound Mix w/ SIM Compounds (18 components)

acenaphthylene-d8	fluoranthene-d10
anthracene-d10	fluorene-d10
benzo(a)pyrene-d12	2-methylnaphthalene-d10
bis(2-chloroethyl)ether-d8	4-methylphenol-d8
4-chloroaniline-d4	nitrobenzene-d5
2-chlorophenol-d4	2-nitrophenol-d4
2,4-dichlorophenol-d3	4-nitrophenol-d4
dimethylphthalate-d6	phenol-d5
4,6-dinitro-2-methylphenol-d	pyrene-d10

2,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 33918 (ea.)

Semivolatiles: Calibration Mixes

SOM01.1 SVOA MegaMix™ (67 components)

acenaphthene	2,4-dinitrophenol
acenaphthylene	2,4-dinitrotoluene
acetophenone	2,6-dinitrotoluene
anthracene	diphenylamine[1]
atrazine	di-*n*-octyl phthalate
benzaldehyde	fluoranthene
benzo(a)anthracene	fluorene
benzo(a)pyrene	hexachlorobenzene
benzo(b)fluoranthene	hexachlorobutadiene
benzo(ghi)perylene	hexachlorocyclopentadiene
benzo(k)fluoranthene	hexachloroethane
biphenyl	indeno(1,2,3-cd)pyrene
bis(2-chloroethoxy)methane	isophorone
bis(2-chloroethyl)ether	2-methylnaphthalene
bis(2-chloroisopropyl) ether	2-methylphenol
bis(2-ethylhexyl)phthalate	3-methylphenol*
4-bromophenyl phenyl ether	4-methylphenol*
butyl benzyl phthalate	naphthalene
ε-caprolactam	2-nitroaniline
carbazole	3-nitroaniline
4-chloro-3-methylphenol	4-nitroaniline
2-chloronaphthalene	nitrobenzene
2-chlorophenol	2-nitrophenol
4-chlorophenyl phenyl ether	4-nitrophenol
chrysene	N-nitroso-di-*n*-propylamine
dibenz(a,h)anthracene	pentachlorophenol
dibenzofuran	phenanthrene
3,3'-dichlorobenzidine	phenol
2,4-dichlorophenol	pyrene
diethyl phthalate	1,2,4,5-tetrachlorobenzene
2,4-dimethylphenol	2,3,4,6-tetrachlorophenol
dimethyl phthalate	2,4,5-trichlorophenol
di-*n*-butyl phthalate	2,4,6-trichlorophenol
4,6-dinitro-2-(dinitro-ocresol)	

1,000µg/mL each in methylene chloride (*3-methylphenol and 4-methylphenol at 500µg/mL), 1mL/ampul
cat. # 33005 (ea.)

[1]N-Nitroso-diphenylamine (listed analyte) decomposes to diphenylamine (mix component).

Semivolatiles: QA Mixes

SV Internal Standard Mix

acenaphthene-d10	naphthalene-d8
chrysene-d12	perylene-d12
1,4-dichlorobenzene-d4	phenanthrene-d10

2,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 31206 (ea.)

4,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 31006 (ea.)

Revised SV Internal Standard Mix (7 components)

acenaphthane-d10	naphthalene-d8
chrysene-d12	perylene-d12
1,4-dichlorobenzene-d4	phenanthrene-d10
1,4-dioxane-d8	

2,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 31885 (ea.)

4,000µg/mL each in methylene chloride, 1mL/ampul
cat. # 31886 (ea.)

SOM01.1 SVOA B/N Matrix Spike Mix

acenaphthene	N-nitroso-di-*n*-propylamine
2,4-dinitrotoluene	pyrene

5,000µg/mL each in methanol, 1mL/ampul
cat. # 33916 (ea.)

5,000µg/mL each in methanol, 5mL/ampul
cat. # 33917 (ea.)

B/N Matrix Spike Mix

acenaphthene	N-nitroso-di-*n*-propylamine
1,4-dichlorobenzene	pyrene
2,4-dinitrotoluene	1,2,4-trichlorobenzene

1,000µg/mL each in methanol, 1mL/ampul
cat. # 31004 (ea.)

5,000µg/mL each in methanol, 1mL/ampul
cat. # 31074 (ea.)

5,000µg/mL each in methanol, 5mL/ampul
cat. # 31084 (ea.)

Acid Matrix Spike Mix

4-chloro-3-methylphenol	pentachlorophenol
2-chlorophenol	phenol
4-nitrophenol	

1,500µg/mL each in methanol, 1mL/ampul
cat. # 31005 (ea.)

7,500µg/mL each in methanol, 1mL/ampul
cat. # 31075 (ea.)

7,500µg/mL each in methanol, 5mL/ampul
cat. # 31085 (ea.)

SV Tuning Compound

decafluorotriphenylphosphine (DFTPP)

2,500µg/mL in methylene chloride, 1mL/ampul
cat. # 31001 (ea.)

Organochlorine Pesticide Resolution Check Mix

(22 components)

aldrin	10µg/mL	endosulfan I	10
α-BHC	10	endosulfan II	20
β-BHC	10	endosulfan sulfate	20
δ-BHC	10	endrin	20
γ-BHC (lindane)	10	endrin aldehyde	20
α-chlordane	10	endrin ketone	20
γ-chlordane	10	heptachlor	10
decachlorobiphenyl	20	heptachlor epoxide	
dieldrin	20	(isomer B)	10
4,4'-DDD	20	methoxychlor	100
4,4'-DDE	20	2,4,5,6-tetrachloro-	
4,4'-DDT	20	*m*-xylene	10

In hexane:toluene, 1mL/ampul
cat. # 32454 (ea.)

Pesticide Surrogate Mix

decachlorobiphenyl	200µg/mL
2,4,5,6-tetrachloro-*m*-xylene	100

In P&T methanol, 1mL/ampul
cat. # 32453 (ea.)

Aroclor® 1016/1260

400µg/mL each in acetone, 1mL/ampul
400µg/mL in acetone, 1mL/ampul
cat. # 32456 (ea.)

for more info

For reference materials for OLC 03.2 analyses of pesticides or Aroclor® PCBs, please visit our website, or call your Restek representative.

free data

Available on Our Website: Lot Certificates, Datapacks, and MSDS

For complete information detailing manufacturing and testing for Restek inventoried reference standards, just visit our website at **www.restek.com** To view lot certificates and/or an MSDS, enter the catalog number of the product in the Search feature. For a free Datapack, enter the catalog number and lot number of the product, to obtain a printable pdf file.

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We have over 2,000 pure, characterized, neat compounds in our inventory! If you do not see the EXACT mixture you need listed on any of these pages, call us.

For our on-line Custom Reference Materials Request Form visit us on the web at **www.restek.com/solutions**.

Improve Characterization of Complex Protein Digests

Using Viva Wide Pore HPLC Columns

by Julie Kowalski, Innovations Chemist

- Superior resolution—many peaks contain one or two peptides, not three or more.
- Excellent results with highly aqueous mobile phases, compatible with digest matrices.
- Restek-manufactured silica in Restek-manufactured columns.

Protein analyses often incorporate a combination of liquid chromatography and electrospray mass spectrometry. Typically, a protein sample is chemically or enzymatically digested to produce peptides, HPLC/MS is used to resolve and identify the peptides, and this information is used to search protein databases to identify the protein of interest. This type of analysis is now used in many fields, including the bioanalytical and pharmaceutical disciplines.

We tailored Viva silica specifically to provide superior chromatography for peptides and other large molecules, and we highly recommend Viva columns for analyses of protein digests. Featuring the largest available surface area in 250-350 Angstrom pores, packings prepared from Viva silica allow longer interaction between peptides and the stationary phase, affording greater resolution.

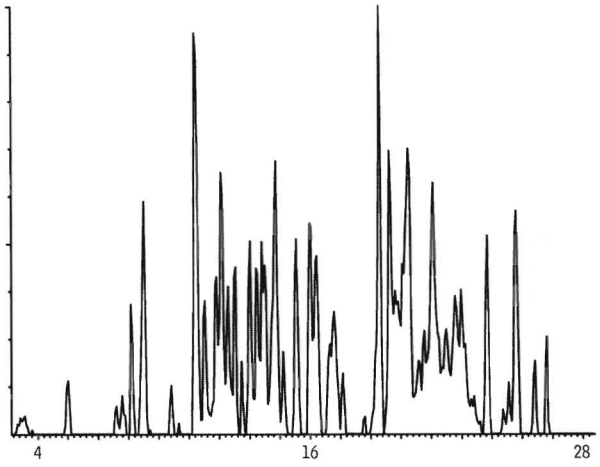
For an example analysis, we prepared a trypsin digest of bovine serum albumin (BSA).[1] We used a 150mm x 1mm ID Viva C18 column (5µm particles, cat# 9514561) to separate the peptides, which number approximately 70, and identified them through manual data analysis.

Figure 1 is a TIC chromatogram for the BSA trypsin digest. Close observation reveals the Viva C18 column has provided outstanding separation, based on the large number of discrete peaks representing only one or two peptides. In contrast, in typical results from other "wide pore" columns it is common to see three or more peptides per peak; this can reduce the number of peptides that are identified. The large number of discrete peaks in Figure 1 also indicates that peptide interaction with the Viva C18 stationary phase, rather than with one another, is the primary retention/separation mechanism.

Figure 1 A Viva C18 column resolves a BSA tryptic digest into many 1-2 peptide peaks, for more reliable identification.

for more info

For details on this analysis, please visit our website: **www.restek.com/bioanalytical**



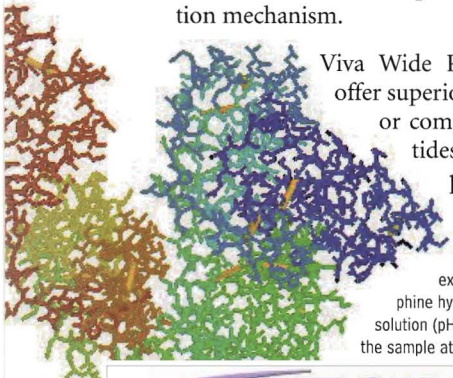
Sample:
Inj.: 15µL
Conc.: bovine serum albumin tryptic digest, 16pmol/µL
Sample diluent: water/0.15% formic acid (v/v)

Column: **Viva C18** (cat. # 9514561)
Dimensions: 150mm x 1mm
Particle size: 5µm
Pore size: 300Å

Conditions:
Mobile Phase: A: water/0.15% formic acid (v/v)
B: acetonitrile/0.15% formic acid (v/v)

Time:	%B
0.0	5
4.5	5
64.0	65

Flow: 0.2mL/min.
Temp.: ambient
Det.: Micromass Quattro II



Viva Wide Pore HPLC Columns offer superior resolution of simple or complex mixtures of peptides - a critical factor in protein identifications.

[1] BSA disulfide bonds were reduced by adding a molar excess of tris(2-carboxyethyl)phosphine hydrochloride (TCEP) to a buffered solution (pH 7) containing BSA. We stored the sample at 40°C for one hour, under argon,

Viva C18 Columns

Physical Characteristics:

particle size: 3µm or 5µm, spherical
pore size: 300Å
carbon load: 9%
endcap: yes
pH range: 2.5 to 10
temperature limit: 80°C

Length	1.0mm ID cat.#	2.1mm ID cat.#	3.2mm ID cat.#	4.6mm ID cat.#
5µm Columns				
30mm	9514531	9514532	9514533	9514535
50mm	9514551	9514552	9514553	9514555
100mm	9514511	9514512	9514513	9514515
150mm	9514561	9514562	9514563	9514565
200mm	9514521	9514522	9514523	9514525
250mm	9514571	9514572	9514573	9514575

Sidewinder™ Column Heaters and Heater/Cooler

By Becky Wittrig, Ph.D., HPLC Product Manager

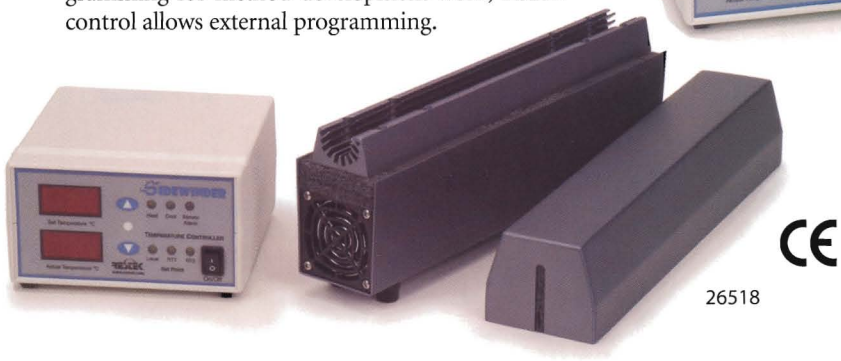
- Easy to set up!
- Lightweight and compact—require little bench space.
- Heaters operate from 5°C above ambient to 85°C, heater/cooler from 5°C to 55°C.

Temperature stability is critical for accurate and reproducible results in HPLC analyses. Sidewinder™ Column Heaters provide optimum heating performance and accuracy to within 1°C. The unique sleeve design completely encloses any analytical HPLC column up to 25cm long. State-of-the-art electronics in the 24V control unit allow fast 10Hz sampling and stability to within 0.1°C. RS232 control allows external programming.

Sidewinder™ Column Heater/Coolers are designed to hold an HPLC column up to 30cm long and 7.8mm in diameter. A doubly insulated cover maintains temperature stability in the chamber to within ±0.2°C. The high performance Peltier-driven 24V control unit allows remote temperature programming for method development work; RS232 control allows external programming.

26516

26517



26518

new and Improved!

Description	qty.	cat.#
Temperature Control Module and Long Column Holder, 25cm Holder	ea.	26516
Temperature Control Module and Short Column Holder, 10cm Holder	ea.	26517
Sidewinder™ Heater/Cooler Temperature Control Module	ea.	26518

All Sidewinder™ temperature control products carry the value recognized CE mark. Each unit meets the demanding electromagnetic emission standards of the new European Union Directives, United States standards, and Canadian standards.

MicroPulse™ Pulse Dampers

- Compact unit (2.5" x 1.5") can be placed almost anywhere.
- Small, 150µL dead volume at atmospheric pressure.
- Compatible with high pressure (stainless steel unit to 6000psi, PEEK® unit to 5000 psi).

The MicroPulse™ pulse damper improves system baseline stability while increasing system volume by only 150µL—ideal for applications in which minimizing total system volume is critical. 316 stainless steel or PEEK® option, for a wide range of applications.

Description	qty.	cat.#
MicroPulse™ Pulse Damper, Stainless Steel	ea.	25238
MicroPulse™ Pulse Damper, PEEK®	ea.	25239

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Parker ChromGas® Hydrogen Generators

Is your lab wasting money on bottled gas?

by Barry Burger, Petroleum Chemist

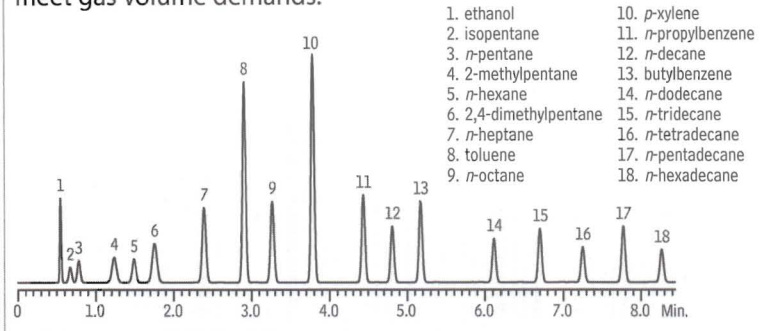
- Economical, continuous source of ultra-pure hydrogen (99.9995%).
- Safe and easy to use and maintain.
- Hydrogen reduces gas costs, cuts analysis time by 50%, increases column lifetimes.

If you use 2-3 cylinders of helium and/or hydrogen per week, as carrier gas and/or fuel gas, bottled gas is an expense in the range of $15,000 to $25,000 per year•, including overhead: expenses and time involved with ordering, transporting, installing, and periodically inspecting cylinders. You also contend with unquantifiable costs, such as floor space lost to an inventory of cylinders. Helium, widely used as carrier gas, is a non-renewable resource extracted from natural gas and, because it is a petrochemical product, its cost will continue to rise, domestically and internationally. Chromatographers must look for cost effective, ultra-pure gas alternatives to supply their instruments and state-of-the-art analytical columns. Fortunately, we do have options.

Past practice in gas chromatography was to select either nitrogen or helium as the carrier gas. Hydrogen wasn't given much consideration, primarily because of flammability and storage issues, even though it offers several distinct advantages over nitrogen or helium. Now, Parker ChromGas® hydrogen generators are a safe, reliable source of ultra-pure (99.9995%) hydrogen, and effective replacements for bottled gas. A Parker ChromGas® hydrogen generator stores less than 50mL of hydrogen (less than 0.002 cubic feet) at 1 atm., or 305mL of hydrogen (0.01 cubic feet) at 6.1 atmospheres (90psig.) From a safety standpoint there is no compromise, compared to a 300 cubic foot cylinder of hydrogen at 2500 psig.

Parker ChromGas® hydrogen generators continuously produce dry, ultra-pure hydrogen by electrolytic dissociation of deionized water and hydrogen proton conduction across a membrane. The hydrogen product is dried by passing it through a coalescing filter, a drying tube, and a desiccant cartridge. Maximum output pressure, 90psig, is controlled to the point of use via a pressure adjust regulator. Other safety features include a pressure relief valve to prevent overpressurization and a

Figure 1 A simulated distillation reference mix is well resolved, in 8 minutes, showing a Parker ChromGas® hydrogen generator can meet gas volume demands.



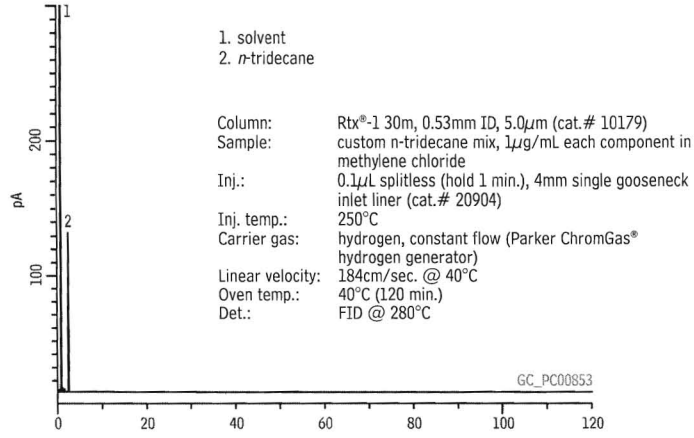
Column: Rtx®-1 30m, 0.53mm ID, 5.0µm (cat.# 10179)
Sample: custom paraffin mix, neat, 2.7-12.9% each component
Inj.: 0.1µL splitless (hold 1 min.), 4mm single gooseneck inlet liner (cat.# 20904)
Inj. temp.: 250°C
Carrier gas: hydrogen, constant flow (Parker ChromGas® hydrogen generator)
Linear velocity: 184cm/sec. @ 40°C
Oven temp.: 40°C (1 min.) to 265°C @ 25°C/min. (hold 4 min.)
Det.: FID @ 280°C

GC_PC00852

Table 1 Repeatable retention times for simulated distillation mix components confirm the hydrogen generator's steady and precise flow of carrier gas.

				Run Number / Retention Time (min.)									
Component	Mean	SD	%RSD	1	2	3	4	5	6	7	8	9	10
1. ethanol	0.547	1E-03	0.1765	0.546	0.548	0.548	0.548	0.549	0.546	0.547	0.547	0.547	0.548
2. isopentane	0.67	1E-03	0.1484	0.669	0.67	0.671	0.67	0.672	0.669	0.669	0.67	0.669	0.67
3. *n* pentane	0.779	0.001	0.169	0.777	0.779	0.78	0.779	0.781	0.777	0.778	0.779	0.778	0.78
4. 2-methylpentane	1.232	0.001	0.1198	1.229	1.231	1.233	1.232	1.234	1.23	1.232	1.232	1.232	1.233
5. *n*-hexane	1.488	0.001	0.0992	1.485	1.487	1.489	1.488	1.49	1.486	1.488	1.488	1.488	1.489
6. 2,4-dimethylpentane	1.753	0.001	0.0721	1.751	1.752	1.754	1.754	1.755	1.752	1.754	1.754	1.754	1.754
7. *n*-heptane	2.387	0.001	0.0442	2.385	2.386	2.388	2.387	2.388	2.386	2.387	2.387	2.388	2.388
8. toluene	2.904	0.001	0.0356	2.902	2.904	2.905	2.904	2.905	2.903	2.904	2.905	2.905	2.905
9. *n*-octane	3.266	7E-04	0.0214	3.264	3.265	3.266	3.266	3.266	3.265	3.266	3.266	3.266	3.266
10. *p*-xylene	3.784	7E-04	0.0195	3.783	3.784	3.785	3.784	3.784	3.783	3.784	3.784	3.785	3.785
11. *n*-propylbenzene	4.438	5E-04	0.0109	4.437	4.438	4.438	4.438	4.438	4.437	4.437	4.438	4.438	4.438
12. *n*-decane	4.809	4E-04	0.0088	4.809	4.809	4.809	4.809	4.809	4.808	4.808	4.809	4.809	4.809
13. butylbenzene	5.174	5E-04	0.0102	5.173	5.174	5.174	5.174	5.173	5.173	5.173	5.173	5.174	5.174
14. *n*-dodecane	6.116	5E-04	0.0079	6.116	6.116	6.116	6.116	6.116	6.115	6.115	6.116	6.116	6.115
15. *n*-tridecane	6.703	5E-04	0.0077	6.704	6.704	6.704	6.704	6.703	6.703	6.703	6.703	6.703	6.703
16. *n*-tetradecane	7.255	7E-04	0.0097	7.256	7.255	7.255	7.255	7.254	7.254	7.254	7.254	7.254	7.254
17. *n*-pentadecane	7.774	6E-04	0.0081	7.775	7.775	7.775	7.774	7.774	7.773	7.774	7.774	7.774	7.774
18. *n*-hexadecane	8.264	6E-04	0.0069	8.265	8.265	8.264	8.264	8.264	8.263	8.264	8.264	8.264	8.264

Figure 2 Carrier gas from a Parker ChromGas® hydrogen generator assures a stable baseline, for sensitive analyses. Performance equivalent to cylinders, at lower cost.



mass leak sensor to indicate hydrogen demand is exceeding instrument capability, in which case the generator will shut down. A low water level and/or poor quality water also will shut down the generator, to prevent damage to the electrolytic cell.

Maintaining the generator is simple. The 4-liter water reservoir may be filled at any time without shutting down the generator, eliminating the downtime associated with changing gas cylinders. At maximum hydrogen demand, the smallest generator will consume one 4-liter tank of deionized water in 8-10 days. The deionizer bags in the water tank should be replaced twice yearly. An LED indicator will illuminate when the desiccant cartridge requires regeneration.

To evaluate performance, we set up a small Parker ChromGas® hydrogen generator (90mL/min. maximum hydrogen output) to supply both carrier gas and fuel gas to an Agilent 6890 GC. We installed a 30 meter x 0.53mm ID x 5µm df Rtx®-1 column (100% polydimethylsiloxane (PDMS) phase, cat.# 10179) in the oven and set analytical parameters as specified in ASTM D-7096-05, a simulated distillation method, but substituted hydrogen for helium as the carrier gas. We used a column flow rate of 40mL/min., in the constant flow mode, which represented the optimum linear velocity for hydrogen. The 40mL/min. carrier gas flow rate, plus a 40mL/min. flow of fuel gas, was 90% of the generator's maximum output capacity, and tested the generator's capability to meet volume demands.

Figure 1 is a chromatogram of the calibration standard used for retention time-boiling point determination and response factor validation in the ASTM method. The components were well resolved and the analysis completed rapidly, in little more than 8 minutes. Reproducible retention times are vital to obtaining accurate initial boiling point (IBP) data. Table 1 shows retention times for the ASTM reference mix components were well within the method specification of ±0.05 minutes per compound, demonstrating the hydrogen generator's ability to maintain a steady and precise flow of carrier gas. Figure 2 monitors FID baseline stability over 2 hours. These figures and data clearly show that a Parker ChromGas® hydrogen generator is a dependable source of ultra-high purity carrier and fuel gas for demanding GC applications.

On average, yearly electricity and maintenance costs for operating a Parker ChromGas® hydrogen generator are approximately $225•. Offsetting the costs of purchasing and operating a generator with the savings made by not using gas cylinders indicates the generator will pay for itself in 1 to 2 years. With numbers like these, can you afford not to consider purchasing a Parker ChromGas® hydrogen generator for your laboratory?

• Cost estimate for USA, in US $.

Parker ChromGas® Hydrogen Generators

- Selectable delivery pressure: 0–100psig.
- High hydrogen purity—99.9995%.
- Greater convenience and safety.

Parker ChromGas® hydrogen generators are certified for laboratory use by Canadian Standards Association (CSA), Underwriters Laboratories (UL), and International Electrotechnical Commission (IEC) 1010.

Hydrogen Purity:	99.9995%
Outlet Port:	1/8" compression
Electrical:	117 Vac/234Vac
Pressure Control:	5 to 20 psig ±0.5%
	20 to 90 psig ±0.2%
Delivery Pressure:	2 to 30 psig ±3%
	30 to 100 psig ±2%
Shipping Weight:	40 lb (18 kg)
Dimensions:	13"H x 15"W x 14"D (33cm x 38cm x 36cm)

Description	Capacity	cat.#
Hydrogen Generator A9090	90cc/min.	22033
Hydrogen Generator A9150	160cc/min.	22034
Hydrogen Generator B9200	250cc/min.	22035
Hydrogen Generator B9400	500cc/min.	22036
Replacement Deionizer Bag (for all models, 2-pk.)		21670
Replacement Desiccant Cartridge (for all models)		21671

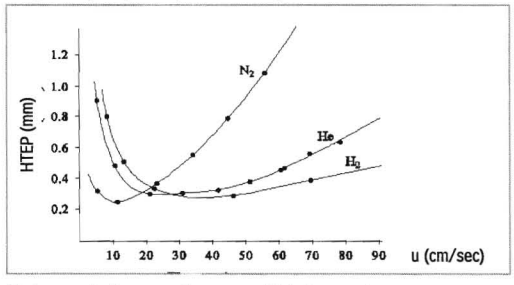
International Power Cord Sets

Location	cat.# suffix
United Kingdom (230VAC, 50/50Hz)	-550
European (230VAC, 50/60Hz)	-551
IEC Connector Only (230VAC, 50/60Hz)	-552
Japanese (200VAC, 50/60Hz)	-556
Japanese for Hydrogen (100VAC, 50/60Hz)	-554

Just add the proper suffix to the catalog number for the gas generator you are ordering.

tech tip

Why use hydrogen as your carrier gas?



Hydrogen, helium, or nitrogen - which do you choose as your carrier gas? We need only look at the van Deemter curves to see the advantages of hydrogen as a carrier gas.Nitrogen generates the highest column efficiency (HETP = 0.22mm), but at an optimum velocity of only 8-10 cm/sec. This great sacrifice in the speed of analysis generally makes nitrogen a poor choice. Column efficiency is slightly reduced with helium (HETP = 0.29mm), but optimum linear velocity is 19-22 cm/sec. With an optimum linear velocity of 35-42cm/sec., hydrogen combines high column efficiency (HETP = 0.28mm) with analysis times 4x faster than nitrogen and 2x faster than helium, thus reducing costs per analysis. Linear velocities of up to 75-80cm/sec. can be used with only a small decrease in column efficiency. Another benefit: lower temperatures are needed to elute analytes, increasing column longevity.

GC Inlet Liner Deactivations for Basic Drug Analysis

By Kristi Sellers, Clinical/Forensic Innovations Chemist, and Lydia Nolan, Innovations Chemist

- Base-deactivated inlet liners are inert to basic drugs, for greater responses.
- Inertness of Rtx®-5Amine column is enhanced for basic compounds.
- Use this liner / column combination for the lowest %RSDs for basic drugs.

Clinical and forensic toxicologists are required to detect low levels of abused drugs in body fluids and confirm their presence by GC/MS. Typical limits of detection are 1-15ng/mL, depending on the sample matrix. For basic drugs (e.g., Figure 1), selecting the proper surface treatment for the GC inlet liner is important, because this parameter can affect responses. The surface of a glass inlet liner contains active silanol groups (Si-OH) that can act as electron pair acceptors, and react with nitrogen or oxygen electron pair donors in basic drug molecules (Figure 2).[1] These reactions usually are rapid and reversible, but they are expressed chromatographically as broad, tailing peaks and/or reduced responses. To eliminate these acid-base reactions, make chromatographic peaks sharp, Gaussian, and easy to integrate, and thereby help ensure reproducible and accurate responses, the -OH groups on the glass surface must be deactivated.

Using GC/FID responses, we evaluated several alternatives for deactivating inlet liners, to determine maximum sensitivity for basic drugs. We prepared reference standards of the free base forms of alprazolam, benzphetamine, cocaine, codeine, ketamine, methadone, and phencyclidine (Figure 1) at 100, 50, 25, 10, and 5 ng/mL concentrations, then analyzed the drugs on a base-deactivated 15m, 0.25mm ID, 0.25µm Rtx®-5Amine column (5% diphenyl/95% dimethylpolysiloxane stationary phase), using a 4mm single gooseneck inlet liner that was untreated, deactivated through an intermediate polarity deactivation process (standard liner deactivation procedure), deactivated through a base deactivation process, or deactivated through the Siltek® deactivation process. We obtained three replicate analyses for each reference standard-liner treatment combination, and evaluated the response data statistically to determine which deactivation treatment maximized sensitivity and reproducibility. We used these results to generate box plots that display the range of data distribution, or variation – an indication of the reproducibility of the performance. We chose phencyclidine (PCP) and cocaine plots to represent the nitrogen-containing and nitrogen/oxygen-containing drugs, respectively (Figure 2). The line in each box indicates the mean response.

Figure 1 Nitrogen- and oxygen-containing compounds can react with silanol groups on glass surfaces, causing poor chromatography.

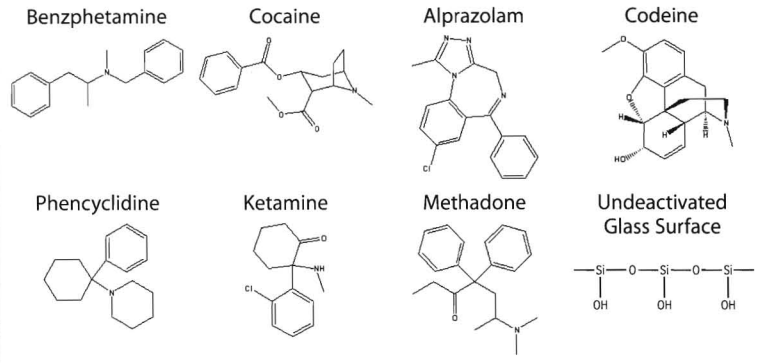


Figure 2 A base-deactivated inlet liner provides highest mean responses for PCP.

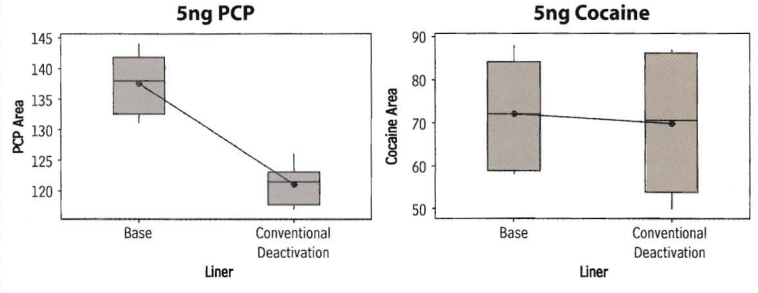
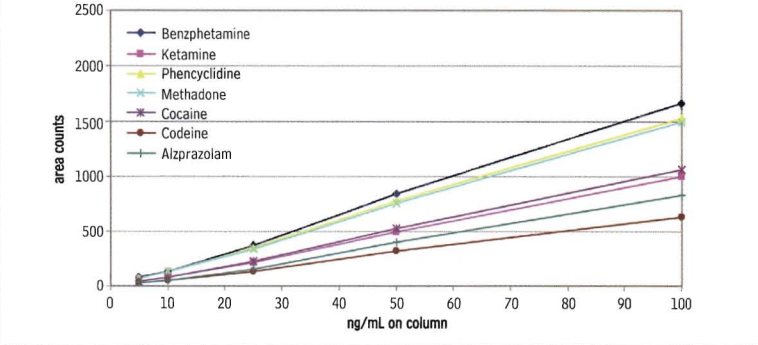


Figure 3 Linearity plots for all drugs, analyzed using a base-deactivated inlet liner and an Rtx®-5Amine column.



Base Deactivated Inlet Liners for Basic Drug Analysis

For Agilent GCs	ea.	cat.# 5-pk.	25-pk.
Gooseneck Splitless (4.0mm ID** x 6.5mm OD x 78.5mm) 	20798-210.1	20799-210.5	20800-210.25
Gooseneck Splitless w/ Base Deactivated Wool (4.0mm ID** x 6.5mm OD x 78.5mm) 	20798-211.1	20799-211.5	20800-211.25
Split Straight w/ Base Deactivated Wool (4.0mm ID** x 6.3mm OD x 78.5mm) 	20781-211.1	20782-211.5	20783-211.25
Cyclosplitter® (4.0mm ID** x 6.3mm OD x 78.5mm) 	20706-210.1	20707-210.5	20708-210.25

**Nominal ID at syringe needle expulsion point.
For liners for other instruments, refer to our catalog or website.

Base-Deactivated Inlet Liners

qty.	Base-Deactivated		Base-Deactivated w/ Base-Deactivated Wool	
each	-210.1	addl. cost	-211.1	addl. cost
5-pk.	-210.5	addl. cost	-211.5	addl. cost
25-pk.	-210.25	addl. cost	-211.25	addl. cost

For base-deactivated inlet liners, add the corresponding suffix number to the liner catalog number.

Base-Deactivated Wool

Ideal for amines and other basic compounds.

Description	qty.	cat.#
Base-Deactivated Wool	10 grams	20999

Mini Wool Puller/Inserter

Insert and remove wool plugs easily.

Description	qty.	cat.#
Mini Wool Puller/Inserter	2-pk.	20114

Not recommended for use with double gooseneck liners.

Inlet Liner Removal Tool

- Easily remove liner from injector—no more burned fingers.
- Made from high-temperature silicone.
- Won't chip or crack the liner.

Description	qty.	cat.#
Inlet Liner Removal Tool	3-pk.	20181

Rtx®-5 Amine Columns (fused silica)

(Crossbond® 5% diphenyl/95% dimethyl polysiloxane)

ID	df (µm)	temp. limits	length	cat. #
0.25mm	0.25	-60 to 300/315°C	30-Meter	12323
0.25mm	0.25	-60 to 300/315°C	15-Meter	12320

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The data show that undeactivated liners and liners that received intermediate polarity treatment provided poorer responses or reproducibility, compared to base-deactivated or Siltek® treated liners, due to the acidic nature of the undeactivated glass surface or to a small but influential number of residual acidic sites remaining on the intermediate polarity deactivated surface.

Because the undeactivated liners and intermediate polarity treated liners exhibited either low mean response or high variation, we reanalyzed the data, excluding these treatments and comparing the remaining data (for base-deactivated liners and Siltek® treated liners) for responses and reproducibility. As shown by the examples in Figure 2, base-deactivated liners and Siltek® treated liners performed equally well for cocaine, but the base-deactivated liners yielded the best responses and reproducibility for PCP. Ultimately, a base-deactivated liner would give the best overall performance. Figure 3 shows the linearity plots for all analyzed drugs, obtained using a base-deactivated liner and an Rtx®-5Amine column. Low %RSD values for ketamine (3%), phencyclidine (2%), methadone (2%), cocaine (3%), codeine (5%), and alprazolam (12%) confirm the reproducibility of data obtained from this combination.

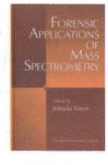
Because nitrogen- and oxygen-containing drugs react with silanol groups on glass surfaces, it is important to use properly deactivated glass inlet liners when analyzing these compounds by GC. This work demonstrates that a base-deactivated inlet liner, used in combination with a base-deactivated column, produces high and reproducible responses for basic drugs.

Reference

1. Seyhan N. and D.C. Ege, *Organic Chemistry* Health and Company, 1984, pp.124-136.

recommended reading

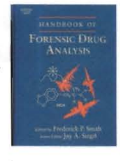
Forensic Applications of Mass Spectrometry



Applies current developments in mass spectrometry to forensic analyses. Techniques discussed include capillary GC/MS, thermospray LC/MS, tandem mass spectrometry, (MS/MS), pyrolysis GC/MS and isotope ratio mass spectrometry.

J. Yinon, CRC Press LLC, 1994, 320pp., ISBN 0-8493-8252-1 **cat.# 23056** (ea.)

Handbook of Forensic Drug Analysis



Provides in-depth, up-to-date methods and results. Chapters by leading researchers discuss the various forms of drugs, as well as the origin and nature of samples.

F. Smith and J. Siegel, Elsevier Academic Press, 2004, 584pp., ISBN 0-12-650641-8 **cat.# 23055** (ea.)

Instrument Innovations!

New Injection Port Can Simplify Life in Your Laboratory

by Donna Lidgett, GC Accessories Product Marketing Manager

- No kinked or broken gas lines.
- Change inlet liners faster, easier, and eliminate touching hot surfaces.
- Excellent for Agilent 5890, 6850, or 6890 GCs; especially advantageous with Agilent GCs equipped with autosamplers.

EZ Twist Top™ Split/Splitless Injection Port for Agilent GCs

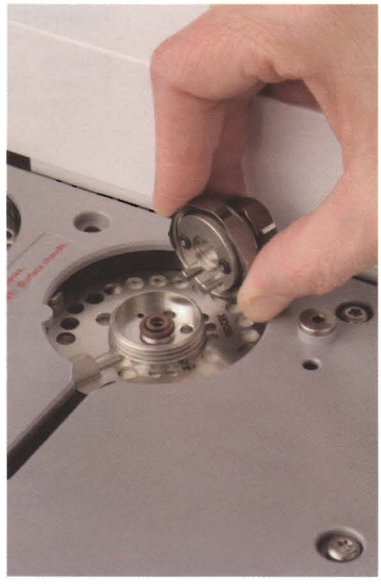
Injection port maintenance should be performed prior to installing any capillary column, and on a routine basis, based on the number of injections made and the cleanliness of the samples. For optimum system performance, the injection port liner must be free of sample residue, septum particles, and ferrule fragments, so proper maintenance includes replacing the injection port liner, critical seals, and septum. Peak shape degradation, poor reproducibility, sample decomposition, and ghost peaks all are associated with using a dirty liner. Frequent septum replacement prevents fragmentation and leaks. Multiple injections and continuous exposure to hot injection port surfaces will decompose the septum and can create particles that can fall into the injection port liner, where they become a potential source of ghost peaks, loss of inertness, and occluded carrier gas flow. Therefore, changing septum and inlet liners frequently is essential to maintaining optimum system performance.

Using Restek's new, unique EZ Twist Top™ Injection Port, and Restek Cool Tools (Septum Nut Removal Tool, cat. # 24918, and Inlet Liner Removal Tool, cat. # 20181—order separately), you can reduce maintenance time and frustration, and eliminate tangled gas lines and damage that leads to leaks, while avoiding direct contact with hot metal and glass surfaces.

The gas lines are attached to the EZ Twist Top™ Shell Weldment (bottom) instead of the weldment (top). Once the injection port is installed the gas lines are under the GC cover and do not interfere with routine injection port maintenance, as shown in Figure 1. To remove the weldment and access the liner, simply slip the Weldment Removal Tool (included in the complete injection port kit) over the weldment (Figure 2), twist, and remove the weldment. For speed and efficiency, the weldment stays secured in the tool until you are ready to reattach it. Changing inlet liners in original equipment injection ports was complicated by the gas lines and sampling tray. Our new injection port makes changing the liner a quick and simple task.

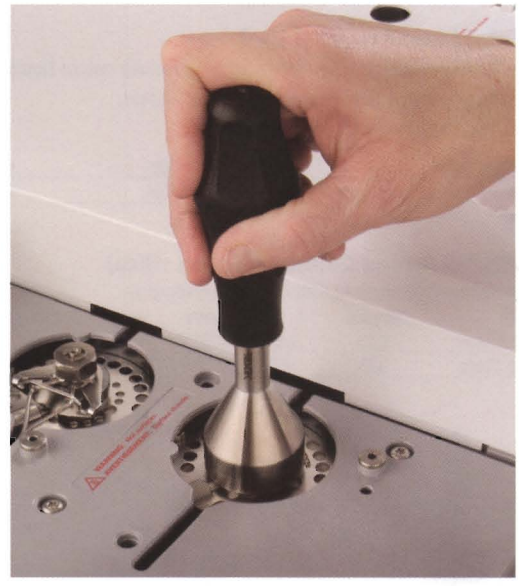
Figure 1

The old way: gas lines can be damaged during routine injection port maintenance.



The new way: with the EZ Twist Top™ Injection Port, the gas lines are under the GC cover and are not disturbed during maintenance.

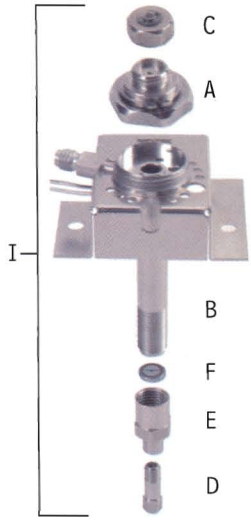
Figure 2



Simply slip the Weldment Removal Tool over the weldment, then twist and remove the weldment. For speed and efficiency, the weldment stays secured in the Weldment Removal Tool until you reattach it.

EZ Twist Top™ Split/Splitless Injection Port for Agilent 5890 GCs

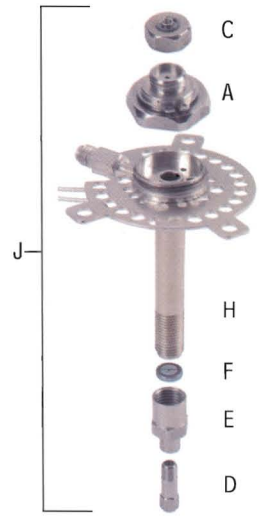
Description	qty.	cat.#
I) **Complete injection port assembly** includes: split/splitless weldment, shell weldment, 2 weldment O-rings, Siltek® Dual Vespel® Ring inlet seal, septum nut, reducing nut, stainless steel capillary nut and weldment tool	kit	22725
Siltek® complete injection port assembly includes: Siltek® split/splitless weldment, Siltek® shell weldment, 2 weldment O-rings, Siltek® Dual Vespel® Ring inlet seal, septum nut, reducing nut, stainless steel capillary nut and weldment tool	kit	22726
A) Split/Splitless Weldment for Agilent 5890/6890/6850 GCs (2 weldment O-rings are installed on the weldment)	ea.	22724
Siltek® Split/Splitless Weldment for Agilent 5890/6890/6850 GCs (2 weldment O-rings are installed on the weldment)	ea.	22732
B) Shell Weldment for Agilent 5890 GCs	ea.	22727
Siltek® Shell Weldment for Agilent 5890 GCs	ea.	22731
Weldment O-rings	10-pk.	22729
C) Autosampler & PTV Septum Nut (for 23-gauge needles)	ea.	20631
D) Stainless Steel Capillary Column Nut (for use with standard 1/16" ferrules)	2-pk.	20883
E) Reducing Nut	ea.	22078
F) Siltek® 0.8mm ID Dual Vespel® Ring Inlet Seal	2-pk. 10-pk.	21242 21243
G) Weldment Removal Tool for Agilent 5890/6890/6850 GCs	ea.	22728



Septum Nut Removal Tool (sold separately)

EZ Twist Top™ Split/Splitless Injection Port for Agilent 6890/6850 GCs

Description	qty.	cat.#
J) **Complete injection port assembly** includes: split/splitless weldment, shell weldment, 2 weldment O-rings, Siltek® Dual Vespel® Ring inlet seal, septum nut, reducing nut, stainless steel capillary nut and weldment tool	kit	22721
Siltek® complete injection port assembly includes: Siltek® split/splitless weldment, Siltek® shell weldment, 2 weldment O-rings, Siltek® Dual Vespel® Ring inlet seal, septum nut, reducing nut, stainless steel capillary nut and weldment tool	kit	22722
A) Split/Splitless Weldment for Agilent 5890/6890/6850 GCs (2 weldment O-rings are installed on the weldment)	ea.	22724
Siltek® Split/Splitless Weldment for Agilent 5890/6890/6850 GCs (2 weldment O-rings are installed on the weldment)	ea.	22732
H) Shell Weldment for Agilent 6890/6850 GCs	ea.	22723
Siltek® Shell Weldment for Agilent 6890/6850 GCs	ea.	22730
Weldment O-rings	10-pk.	22729
C) Autosampler & PTV Septum Nut (for 23-gauge needles)	ea.	20631
D) Stainless Steel Capillary Column Nut (for use with standard 1/16" ferrules)	2-pk.	20883
E) Reducing Nut	ea.	22078
F) Siltek® 0.8mm ID Dual Vespel® Ring Inlet Seal	2-pk. 10-pk.	21242 21243
G) Weldment Removal Tool for Agilent 5890/6890/6850 GCs	ea.	22728



EZ Twist Top™ Split/Splitless Injection Port with Optional Split Vent for Agilent 6890/6850 GCs

Description	qty.	cat.#
Complete injection port assembly includes: split/splitless weldment, shell weldment, 2 weldment O-rings, Siltek® Dual Vespel® Ring inlet seal, septum nut, reducing nut, stainless steel capillary nut and weldment tool	kit	22735
Siltek® complete injection port assembly includes: Siltek® split/splitless weldment, Siltek® shell weldment, 2 weldment O-rings, Siltek® Dual Vespel® Ring inlet seal, septum nut, reducing nut, stainless steel capillary nut and weldment tool	kit	22736
Optional Split/Splitless Shell Weldment (for use with large canister type filter)	ea.	22733
Siltek® Optional Split/Splitless Shell Weldment (for use with large canister type filter)	ea.	22734

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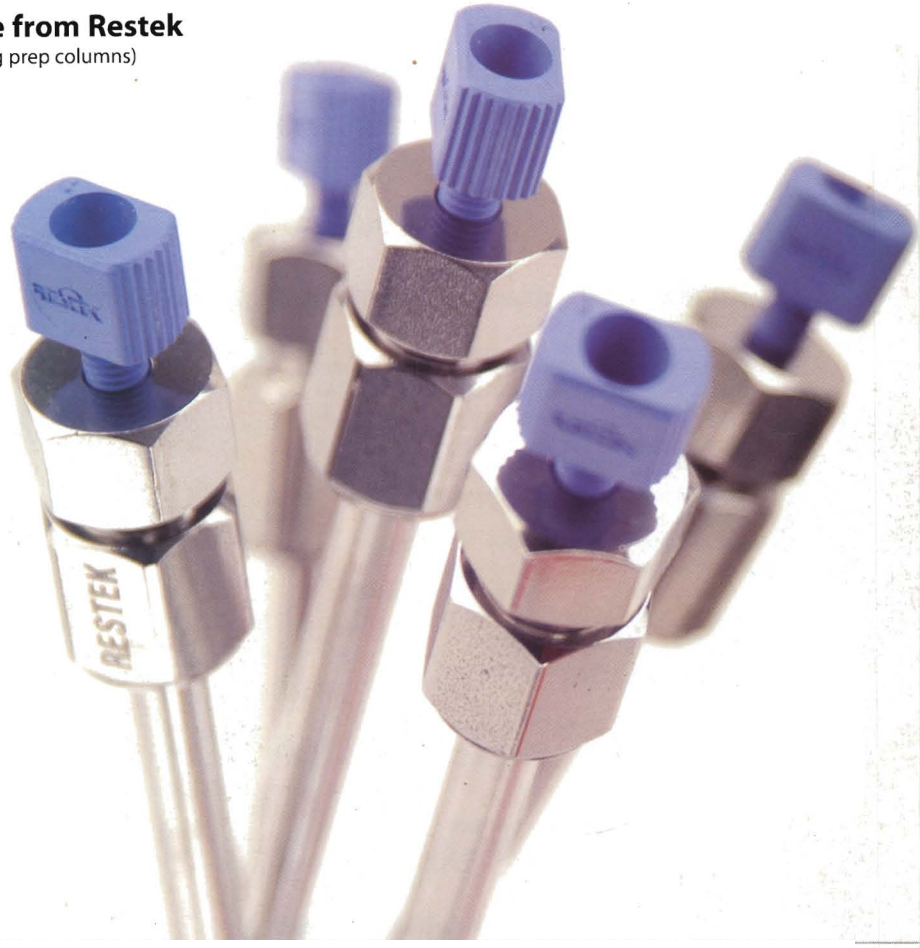
- chiral phases
- bulk packings



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