



COLUMN WATCH

New Chromatography Columns and Accessories at the 2006 Pittcon Conference: Part I

This month's installment of "Column Watch" is the first of a two-part series in which Ron Majors examines the trends and highlights in columns and consumables at Pittcon 2006. Here, he discusses new series of HPLC columns and packings and covers reversed-phase, specialty, adsorption, size-exclusion, and preparative chromatography columns.

Pittcon 2006 — the 57th Pittsburgh Conference on Analytical Chemistry and Applied Spectroscopy — will return to the Orange County Convention Center in Orlando, Florida on March 12–17, 2006. This year's event will host more than 1000 instrument manufacturers and laboratory suppliers in more than 2300 booths. In addition to attending the exposition, the conferees will have the opportunity to listen to more than 2200 oral and poster presentations, which will include invited and award symposia, check out 50 seminar rooms, or attend one of 100 short courses.

Undoubtedly, Pittcon still remains the most important yearly international analytical exhibition where companies introduce their latest instruments, instrument accessories, columns, sample preparation products, and other consumable products. Because many past attendees have purchased one or more new products within three months after attending the show, most exhibitors attempt to maximize their booth traffic to meet as many potential customers as possible.

The purpose of this report is to provide information about many of the new separation consumables and accessory products that will be displayed at Pittcon 2006. In some cases, products that were introduced during 2005 but after Pittcon 2005 (1,2) have been included for reasons of completeness. The information is based upon manufacturers' responses to a questionnaire mailed in December 2005. Because of space limitations and the fact that some manufacturers did not respond to the questionnaire, this report cannot be considered an

exhaustive listing of all new products that will be introduced in Orlando. However, over the years, these Pittcon introduction summaries have provided a good source of information that would be difficult for one individual to gather during the four days of the exhibition. In addition, the products introduced have shown definite correlations to current research, development, and application activity in the separation sciences.

As in previous years, columns and other products recommended by their manufacturers primarily for biomolecule separations or sample preparation are denoted in the tables with the designation BIO. Some of these products can be used for general high performance liquid chromatography (HPLC) separations as well, but their main emphasis is for biological samples. I will cite specific information about bioapplications where appropriate. Because HPLC columns for microbore (1.0–2.1 mm i.d.) and high-throughput HPLC (lengths less than 50 mm) are now numerous, they will be covered in the individual modes.

In this month's coverage, I describe new introductions in the areas of high performance liquid, reversed-phase, normal- and bonded-phase, ion-exchange and ion, size-exclusion (SEC), large- and preparative-scale, and specialty chromatography columns. Next month, I will look at gas chromatography (GC) columns, sample preparation products, and hardware, accessories and kits for chromatography and sample preparation.

Trends and Highlights

General: This year, I observed that there was a continued trend in miniaturization

Companies Listed in This Column

Agela Technologies, Wilmington, Delaware
 Agilent Technologies, Inc., Wilmington, Delaware
 Alltech Associates, Inc., Deerfield, Illinois
 BIA Separations (Astec), Whippany, New Jersey
 BioChrom Labs, Inc., Terre Haute, Indiana
 Bischoff Chromatography, Leonberg, Germany
 Chiral Technologies, Inc., Exton, Pennsylvania
 Daiso Co. Ltd., Osaka, Japan
 Dionex Corp., Sunnyvale, California
 Dynamic Adsorbents, Atlanta, Georgia
 ES Industries, West Berlin, New Jersey
 Fortis Technologies, Neston, United Kingdom
 Hamilton, Reno, Nevada
 ISCO/Teledyne, Mason, Ohio
 Macherey-Nagel GmbH & Co. KG, Düren, Germany
 Merck KGaA, Darmstadt, Germany
 MicroSolv Technology Corp., Long Branch, New Jersey
 OraChrom, Woburn, Massachusetts
 OroChem, Lombard, Illinois
 Restek Corp., Bellefonte, Pennsylvania
 Sepax Technologies, Newark, Delaware
 Supelco, Bellefonte, Pennsylvania
 Thermo Electron Corp., Runcorn, United Kingdom
 Tosoh Bioscience LLC, Montgomeryville, Pennsylvania
 Waters, Milford, Massachusetts
 YMC, Dinslaken, Germany
 ZirChrom Separations Inc., Anoka, Minnesota

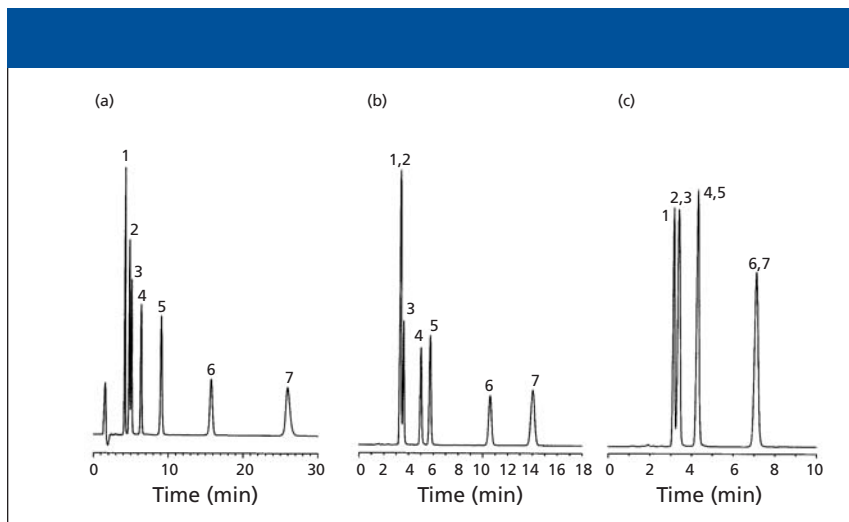


Figure 1: Separation of corticosteroids using (a) an Allure Biphenyl column, (b) a C18 phase, and (c) a phenyl phase. Mobile phase: 60:40 water–acetonitrile; flow rate: 1 mL/min; temperature: ambient. Peaks: 1 = hydrocortisone, 2 = prednisone, 3 = cortisone, 4 = dexamethasone, 5 = corticosterone, 6 = cortisone acetate, 7 = desoxycorticosterone. (Courtesy of Restek Corporation.)

of the chromatographic experiment, not only with the preparation of smaller samples in the microliter-to-submicroliter range but also in the dimensions of the columns, especially in the increasing use of short, sub-2- μm d_p columns in HPLC and LC–mass spectrometry (MS). Still, a large number of products will be introduced for the separation, sample preparation and for LC–MS measurement of biomolecules, still the hottest area in chromatographic research and applications. New breeds of low-bleed phases and ultra inert surfaces for both GC and LC–MS were noted. Monolith technology continues to see new entries of both polymeric- and silica-based stationary

phases.

HPLC columns: Reversed-phase HPLC maintains its dominance and a significant number of regular and specialty reversed-phase columns will be introduced. Particles and column internal diameter (i.d.) are getting smaller and columns are getting shorter with sub-2- μm particles now being offered by several companies; the smallest particle had a diameter of 1 μm . No irregularly shaped silica gel packed columns in analytical sizes will be introduced this year; spherical packings now dominate. Flash chromatography, preparative and process columns, and the availability of bulk packings for self-packing of these

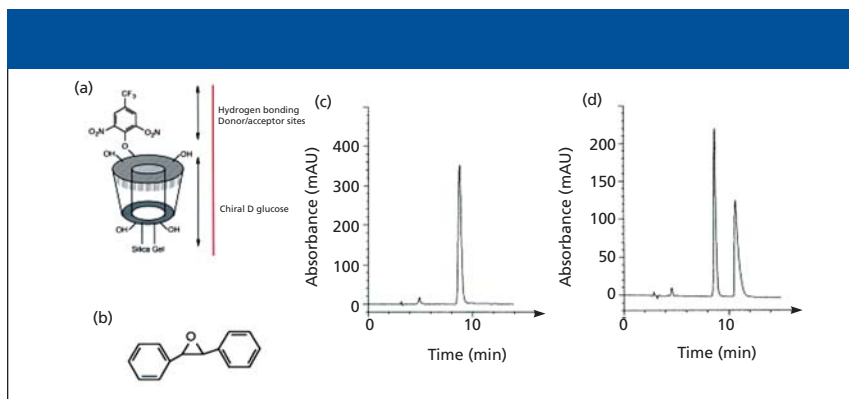


Figure 2: Separation of *trans*-stilbene oxide. Shown are structures of (a) Cyclobond I 2000 DNP and (b) *trans*-stilbene oxide and chromatograms from *trans*-stilbene oxide separations using (c) Cyclobond I 2000 and (d) Cyclobond I 2000 DNP columns. Mobile phase: (c) 25:75 0.1% acetonitrile–TEAA (pH 4.1), (d) 40:60 acetonitrile–0.1% TEAA (pH 4.1); flow rate: 1.0 mL/min; temperature: 25 °C; detection: UV absorbance at 230 nm. (Courtesy of Astec.)

Table I: New series of HPLC columns at Pittcon 2006					
Product Name	Supplier	Analytical	Prep	Guard	Bulk
Dynamic Adsorbents	Dynamic Adsorbents	No	No	No	Yes
GP Series	Sepax Technologies	Yes	No	Yes	No
Hypersil GOLD	Thermo Electron	Yes	Yes	Yes	No
Microbore LC columns	Grace-Alltech	Yes	No	No	No
POPLC	Bischoff Chromatography	Yes	No	No	No
ProntoSIL 90-5	Bischoff Chromatography	Yes	No	No	No
RediSep Purification Kit	Isco/Teledyne	Yes	Yes	No	Yes
Xbridge	Waters	Yes	Yes	Yes	Yes
Zorbax Rapid Resolution HT Extended Range	Agilent Technologies	Yes	No	No	No

columns seem to have had a renaissance, undoubtedly driven by the need to purify new drug candidates and biomolecules. Specialty columns will be numerous with one-third of them recommended for protein-peptide separations and one-third for chiral separations. Columns for hydrophilic interaction chromatography (HILIC) are to be introduced. Carbon nanotube separation columns will be introduced by two companies, one by SEC and one using ion exchange.

Gas chromatography: GC columns

this year will be highlighted with more introductions of lower bleed phases and ultra inert phases that provide enhanced sensitivity, lower baseline drift during temperature programming and improved MS capability. Some have higher temperature limits. A extremely polar phase with 90% biscyanopropyl functionality will be introduced as a truly orthogonal phase for 2-D GC. As usual, several application-specific columns will be introduced, especially for use in the separation of environmental compounds such as pesticides and polychlorinated

biphenyls. In addition, columns for *cis-trans* fatty acids and enantiomers are noted. Only one porous layer open tubular (PLOT) column for oxygenate analysis will be introduced; the rest are wall coated open tubular (WCOT) columns.

Sample preparation: Products for the preparation of tiny amounts of samples in the microliter and submicroliter range will be quite prevalent. Formats range from customizable 96-well solid-phase extraction (SPE) plates and plates with bonded phase surfaces to SPE pipette tips. Many polymeric SPE packings and

Modes	Phases	Comments
Reversed and normal phase hydrophobic and hydrophilic interaction	Silica, alumina, zirconia, titania, zeolites; CN, C2, C8, C18	Variety of packing materials available from 5 μm to 500 μm ; various pore sizes and phase coverages.
Reversed phase	C2, C4, C8, C18	Expanded line of particle and column dimensions to nano and capillary sizes; 50, 75, and 150 μm in addition to microbore and analytical; lengths: 3, 5, 10, and 15 cm; stainless steel (SS) construction; monomeric bonding and endcapped; pore size: 120 \AA ; particle size: 2 μm .
Reversed phase	C8, AQ, CN, PFP	Product line based on high purity silica base material with various bond phase chemistries; particle sizes: 1.9, 3, 5, 8, and 12 μm ; phase coverage: C8 (8% Carbon); AQ (12% C); CN (4% C); Pentafluorophenyl (PFP) (8% C); pore size: 175 \AA .
Reversed and normal phase, hydrophilic interaction	Various	New low-volume column format from nanobore to capillary to microbore 75–1000 μm ; 50–250 mm length; materials of construction: SS, PEEK, and fused silica; particle sizes: 1.5, 3, and 5 μm ; pore sizes: 100–300 \AA ; zero dead volume "in-line" guard and trap columns; variety of company's packing materials.
Reversed phase	Phenyl, CN, C18, Polar-embedded C18	POPLC = phase optimized liquid chromatography; recommended column set consists of three or more different stationary phases (phenyl, CN, C18, polar-embedded C18) combined with isocratic elution to scout stationary phases for optimum; advantages: no gradient reequilibration; reusable mobile phase, constant detector background, less requirements for LC instrumentation, simple optimization via software, can be used for all forms of chromatography; cartridge columns 20, 40, and 60 mm \times 3.0 mm; segmented so can be coupled in series for mixed-mode separations.
Reversed and normal phase	Phenyl, CN, C18 (SH), Polar-embedded C18 (ace EPS)	Series based on 90 \AA , 5- μm silica base material using polyfunctional bonding and endcapping; coverage: Phenyl (12% Carbon); CN (6% C); C18 (21% C); C18 ace EPS (22.5% C); column dimensions: 10, 20, 40, 60, 80 mm \times 3.0 mm.
Reversed and normal phase, ion exchange	CN, NH_2 , Al_2O_3 , SCX	Kit contains chromatography Flash columns along with TLC plates with matching phases recommended for purifications in drug discovery, especially basic compounds; column sizes range from 4 to 330 g for milligram to multigram purification; TLC plates for method development.
Reversed phase	C6/phenyl, C8, C18, C18 polar-embedded (carbamate)	Silica-organic hybrid particles with 2.5-, 3.5-, 5-, and 10- μm particle sizes; pore size: 135 \AA ; coverage: C6/phenyl = 15% Carbon (3.0- $\mu\text{mol}/\text{m}^2$); C8 = 13% C (3.2- $\mu\text{mol}/\text{m}^2$); C18 = 18% C (3.10 $\mu\text{mol}/\text{m}^2$); C18 polar embedded = 17% C (3.3- $\mu\text{mol}/\text{m}^2$); pH range: 1–12 polar embedded 2–11; alternate selectivities, recommended for method development; same phases as Acquity BEH 1.7 μm allowing transfer for methods among column types.
Reversed and normal phase	SB-C18, -C8, -CN; Eclipse XDB- C8, -C18; Extend-C18	HT = high throughput; high resolution product line extensions with high pressure capability; up to 1000 bar pressure rating; SB = StableBond; XDB = Eclipse Extra Densely Bonded; Extend has high pH capability to 12; particle size: 1.8 μm (scalable to 3.5- and 5.0- μm columns); column dimensions: 1.0-, 2.1-, 3.0-, and 4.6-mm i.d.; 20-, 30-, 50-, 100-, and 150-mm lengths; synthesis-controlled particle size distribution gives lower pressure drop than expected for particle size; temperatures up to 100 $^\circ\text{C}$.

the first molecular-imprinted polymer standard product for chloramphenicol will be introduced this year. Products for the removal of high abundance proteins from serum, mostly based upon affinity resins will be a particularly hot area.

Accessories: I noted 31 new accessories for LC, GC, capillary electrophoresis (CE) and capillary electrochromatography (CEC), and sample preparation. Introductions of new general-purpose laboratory products like vials, caps, fittings, and ferrules will be very prominent this year. In GC, new designs of inlet lin-

ers and unions, a variable flow splitter, and space saving gas generators are noteworthy. A portable hydrogen storage unit should be popular for GC field testing. For LC, column-switching kits and miniaturized and high-pressure hardware systems with low dead volume, and an in-line mobile phase heater-cooler should help high-temperature HPLC performance.

New Series of HPLC Columns and Packings

Each year manufacturers introduce entire

new families of HPLC columns at Pittcon. For the sake of brevity, I have elected to cover those series here instead of listing each column under individual headings in the modes' tables. Table I covers those new families of HPLC columns and packings displayed at Pittcon 2006. In some cases, the series are major extensions of existing products in the companies' offerings but covering an entire mode or group of phases while in other cases the series are entirely new to the market.

The highlight of new families of prod-

Table II: Reversed-phase chromatography columns at Pittcon 2006

Product Name	Supplier	Type	Base Material	Functional Group	Particle Size (μm)
Allure Biphenyl	Restek Corp.	Regular	Silica	Biphenyl	3, 5
Ascentis C8	Supelco	Regular	Silica	C8	3, 5
Epic PE	ES Industries	Polar embedded	Silica	Polar embedded C18	3 to preparative
Fortis C18	Fortis Technologies	Regular	Silica	C18	3, 5, 10
Fortis H2o	Fortis Technologies	High aqueous	Silica	C18	3, 5, 10
Nucleodur C18 Isis	Macherey-Nagel	High coverage	Silica	C18	3, 5
PHidelity C18	Restek Corp.	Regular, high pH	Silica	C18	3
ProntoSIL 200-5-C30	Bischoff Chromatography	High coverage	Silica	C30	5
PRP-Plus	Hamilton	Regular	Polymeric	C18	3-75
Quantum 120 A	OroChem	Regular	Silica	C8, C18, CN	5
SunFire C8 and C18	Waters	Regular	Silica	C8, C18	2.5
TSKgel ODS-100V	Tosoh Corp/Tosoh Bioscience	High aqueous	Silica	C18	5
TSKgel ODS-100Z	Tosoh Corp/Tosoh Bioscience	Regular	Silica	C18	5
Venusil ASB-C18	Agela Technologies	High aqueous	Silica	C18	3, 5
Venusil Dual	Agela Technologies	High aqueous	Silica	Polar embedded C18	5
YMC Ultra Fast	YMC	Regular	Silica	C18	2.0

ucts was the introduction of sub-2 μm packings and columns by five companies. Small particles packed into short columns give high throughput separations without loss of resolution or when packed into long columns give high resolution separation of more complex mixtures, albeit at the higher operating pressure. Agilent Technologies (Santa Clara,

California), Grace-Alltech (Deerfield, Illinois), Sepax Technologies (Newark, Delaware), ThermoElectron (Bellefonte, Pennsylvania), and Waters (Milford, Massachusetts) all had small particle columns. These small particles were packed into columns as short as 20 mm and as long as 150 mm and internal diameters as small as 0.075 mm and as

large as 4.6 mm. In these small particle columns, a wider variety of stationary phases are available this year compared to Pittcon 2005 (1).

Flash chromatography packings and columns were also introduced. Flash chromatography has been made more convenient by the introduction of prepacked columns and dedicated instru-

Pore Size (Å)	Comments
60	Highly selective phase for unsaturated compounds; available in microbore-to-preparative dimensions (30–300 mm × 1.0–50 mm); guard columns available.
100	Highly retentive C8 phase; coverage: 15% carbon; recommended for gradient applications, general applications column; column dimensions: microbore, analytical and preparative; guard columns available.
Unspecified	High density polar embedded phase; shows high loading capacity; recommended for the separation of polar compounds; unique selectivity relative to C18; nano to preparative column dimensions; cartridge design; guard column and bulk packing available.
Unspecified	Hydrophobic phase; phase coverage: 17% carbon; pH range 1–11; general phase for reversed-phase applications; microbore to preparative column dimensions; guard columns available.
Unspecified	Hydrophobic phase also shows hydrophilic interactions due to polar endcapping; phase coverage: 18% carbon; enhanced retention of polar analytes under highly aqueous conditions; recommended for basic, acidic, and neutral compounds; microbore to preparative column dimensions; guard columns available.
110	Densely coated phase; 20% carbon coverage; pH range: 1–10; provide hydrophobic and π - π interactions as well as good steric selectivity; wide variety of column dimensions ranging from capillary to preparative; guard columns available.
140	Stable from pH 1–12; temperature limit: 200 °C; polycarbosilane barrier layer protects the silica particle; second layer with C18 attached; dimensions: 30–150 mm × 2.1 and 4.6 mm; guard columns available.
200	Long alkyl chain with polyfunctional bonding; phase coverage: 20.5% carbon; very stable and retentive phase with high loadability; general application column; dimensions: 10–80 mm × 3.0 mm.
100	Recommended for high resolution preparative chromatography; small particle size gives lower pressure characteristics than expected; microbore to preparative column dimensions; hardware: cartridge design; guard columns and bulk packing available.
120	Recommended for analysis of small molecules; based on high purity silica; useful in peptide analysis using trifluoroacetic acid eluents at pH less than 2; 87,000 plates/m for 5- μ m C18 packing .
100	Particle size extension to SunFire product line; adds 2.5 μ m to existing 3.5-, 5-, and 10- μ m sizes; higher efficiency and reduced analysis times; high purity silica; phase coverage: C8 (11.5%); C18 (16%); column dimensions: nano (75- μ m) to analytical (4.6 mm) internal diameters.
100	Silica gel (450 m ² /g surface area) incompletely derivatized with difunctional C18 silane to create low density bonded phase; 15% carbon coverage; double endcapping reduces number of accessible silanol groups; improved peak shape for basic and chelating compounds; analytical-size and guard columns available.
100	Silica gel (450 m ² /g surface area) derivatized with difunctional C18 silane to create high density monolayer phase; 20% carbon coverage; endcapping reduces number of accessible silanol groups; stable retention when operating in 95–100% aqueous conditions; recommended for basic drugs and low molecular weight organic acids; polar compounds retained longer than 100Z column; analytical-size and guard columns available.
150	Nonendcapped AQ phase; 100% water compatible; phase coverage: 10% carbon; recommended for analysis of polar and semipolar compounds in drug discovery and metabolite research; stable at low pH; larger pore with reasonable retention.
100	Column can be used in reversed- and normal-phase modes or mixed modes; 18% carbon coverage; recommended for polar compounds and as replacement of some ion-pair methods; improved retentivity for hydrophilic compounds; improved loading capacity for basic compounds; capillary to preparative dimensions; guard columns available.
120	Short columns with small ultrapure silica particles designed for ultrafast LC; same phase as for larger particle YMC C18 so can "downsize" methods to high speed format; phase coverage: 18% carbon; column dimensions: 20–50 mm × 2.0 mm; cartridge design.

ments. Formerly, chemists would pack their own glass columns with large particle silica gel or soft organic gels and by using gravity-fed systems collect fractions from synthesized crude organic or protein mixtures. Now, there almost as many flash chromatography phases available as there are HPLC stationary phases. For those preparing large axial-compress-

sion preparative columns, bulk materials can be purchased and packed or repacked as needed.

The POPLC system was introduced by Bischoff Chromatography (Leonberg, Germany). This is an optimized set of columns with different complementary stationary phases that is used for method development by isocratic mobile phase

scouting rather than by using gradient elution. Their hardware configurations allow them to quickly connect columns in series for mixed separation modes.

Reversed-Phase Chromatography

As usual, many new reversed-phase columns will be introduced at Pittcon

Table III: HPLC Specialty Columns at Pittcon 2006					
Product Name	Supplier	Type	Applications	Base Material	Functional Group
Acclaim	Dionex Corp.	Application-specific	Explosives	Silica	Proprietary
Acquity BEH130 & BEH300 C18	Waters	Application-specific	Peptide mapping	Silica-organic hybrid	C18
Chiralpak Microflow Capillary	Chiral Technologies	Chiral	Enantiomer separation	Silica	Various polysaccharides
Chiralpak QD-AX & QN-AX	Chiral Technologies	Chiral	Enantiomer separation	Silica	0-9-(<i>tert</i> -butylcarbamoyl)-quinidine or -quinine
Chiralpak SFC	Chiral Technologies	Chiral	Enantiomer separation	Silica	Various polysaccharides
Cogent MS + HT	Microsol Technology	High-Throughput LC-MS	High speed, LC-MS	Silica	C18
Cogent UDA	Microsol Technology	Aqueous normal phase	Polar compounds, peptides	Silica	-CH ₂ ⁻ , COOH
Cyclobond I 2000 DNP	Astec	Chiral	Enantiomer separation	Silica	See description
Cyclobond I 2000 HP-RSP	Astec	Chiral	Enantiomer separation	Silica	Hydroxypropyl β -cyclodextrin
Daisogel SP-300 BIO	Daiso Co.	Application-specific	Proteins and peptides	Silica	C4, C8, C18
Epic HILIC-HC	ES Industries	HILIC	Polar compounds	Silica	Bonded hydroxyl groups
Epic HILIC-RP	ES Industries	HILIC	Polar and nonpolar compounds	Silica	Hydroxyl groups and C18
Hydrocell NS 1500	BioChrom Labs	Application-specific	Carbon nanotubes	Polystyrene-divinylbenzene	Quaternary amine
MRP-C18	Agilent	Application-specific	Proteins	Silica	C18
P-CAP-DP	Astec	Chiral	Enantiomer separation	Silica	<i>R,R</i> - or <i>S,S</i> -phenyldiamine aryl
ProPac IMAC-10	Dionex Corp.	Application-specific	Metal-binding proteins	Ethylvinylbenzene-divinylbenzene	Iminodiacetate
ProSwift	Dionex Corp.	Monolith	Large biomolecules	Polystyrene-divinylbenzene Polymethacrylate	Phenyl and quaternary amine
PRP-Derivative	Hamilton	Application-specific	Oligonucleotide Purification	Polymeric	Proprietary

d_p (μm)	Pore Size (\AA)	Comments
5	120	Recommended for the baseline separation of the 14 explosives covered in the EPA 8330 method; can also separate other important explosives including PETN, EGDN, TATB, and trimethylolethane trinitrate; complementary column to current primary ODS column for EPA 8330; matrices can be soil, ground-, surface- and wastewater.; column dimensions: 250 mm \times 4.6 mm (analytical) and 10 mm \times 4.3 mm (guard); surface area: 300 m ² /g; pore volume: 0.9 mL/g.
1.7	130, 300	BEH= bridged ethyl hybrid; recommended for peptide mapping using using formic or trifluoroacetic acid-acetonitrile gradients; designed and QC tested with actual protein tryptic digest to ensure column-to-column performance; better resolution and sensitivity than traditional peptide mapping HPLC columns; phase coverage: 18% carbon (3.1 $\mu\text{mol}/\text{m}^2$). BIO.
5	Unspecified	Expansion of polysaccharide chiral phase line to cover capillary dimensions; recommended for sample limited situations; can be run up to four times faster than conventional chiral columns while reducing solvent usage; available in AD-H, AS-H, OD-H, and OJ-H phases; column dimensions: 150 mm \times 0.3 mm.
5	Unspecified	Recommended for chiral acids in reversed-phase and polar organic modes; QN-AX gives reversed elution order compared with QD-AX column; complements polysaccharide phases also offered; phases developed by Wolfgang Lindner, University of Vienna; column dimensions: 150 mm \times 2.1, 4.6, and 20 mm; guard columns available.
5	Unspecified	Expansion of polysaccharide chiral phase line to cover analytical and preparative SFC; phases include 1A, 1B, AD-H, AS-H, OC-H, and OJ-H phases; column dimensions: 100–250 mm \times 4.6, 10, 20, 30, and 50 mm.
3, 5	Unspecified	Base-deactivated, low trace metal silica; column dimensions: 50 mm \times 2.0 mm; 16% carbon coverage.
4	100	Column can be used with high organic to retain polar compounds enhancing mass spectrometric sensitivity; stationary phase terminates in Si-H groups instead of Si-OH; phase can be used for reversed phase, normal phase and aqueous normal phase modes; column dimensions from nano to preparative dimensions; guard columns available.
5, 10	100	Phase: 2,6-dinitro-4-trifluoromethyl phenyl ether β -cyclodextrin; broad-based chiral selector with enhanced resolution and selectivity over conventional β - and derivatized-cyclodextrins predominantly for reversed-phase mode; microbore to preparative dimensions; guard columns available.
5, 10, 16	100	Recommended for basic and neutral racemates; increased selectivity for antifungal agents, Jacobsen's catalyst and a number of primary amines: recommended for reversed-phase and polar organic modes; can use MS-compatible mobile phases; microbore to preparative dimensions; guard columns available.
5, 10, 15, 20	300	Available in bulk only; high phase coverage provides strong acid and base resistance in reversed-phase applications. BIO.
3 to preparative	Unspecified	HILIC column for use with organic solvents; high loading capacity; nano to preparative dimensions; cartridge design; guard columns and bulk packing available.
3 to preparative	Unspecified	Dual HILIC column with normal and reversed phase solvent capability; nano to preparative dimensions; cartridge design; guard columns and bulk packing available.
10	500	Recommended for separating metallic from semiconducting-carbon nanotubes (CNT) as well as their diameter-dependent separation; a new application for anion-exchange nucleic acid column; separation appears to be dependent on oligonucleotide sequence; stainless steel column dimensions: 75 mm \times 7.8 mm. BIO
5	Macroporous	Reversed-phase column is especially suited for high recovery (90–95%) for proteins, membrane protein, and lipid separation from biological samples; column dimensions: 50 mm \times 4.6 mm, custom sizes available upon request; BIO.
opolymer	5	Unspecified
	5	Unspecified
	10	Nonporous
	10	An immobilized metal affinity chromatography (IMAC) column for use in separating his-tagged proteins, phosphopeptides, prion peptides, and monoclonal antibodies; additionally can be used for purification, polishing, and for on-column refolding studies; has surface bound nanoparticles; microbore (1- and 2-mm i.d.), analytical (4 mm) and preparative (9 and 22 mm) column dimensions. BIO.
	Not applicable	Unspecified
	Not applicable	Mixed-mode reversed-phase and anion-exchange column; recommended for high throughput, high resolution separation of biomolecules; monolith design allows high flow rate with low back pressure; can be used of LC-MS separations, for desalting and preconcentration of protein samples; dimensions: 50 mm \times 4.6 mm; Stainless steel and PEEK construction; BIO.
	3–75	Unspecified
	3–75	Derivatized polymer is highly selective for purification oligonucleotides; significantly higher yields because it releases purified oligos in final step; microbore to preparative dimensions available; cartridge design; guard columns and bulk packing available; BIO.

Table III: HPLC Specialty Columns at Pittcon 2006 continued

Product Name	Supplier	Type	Applications	Base Material	Functional Group
Styros Benzamidine	OraChrom	Affinity	Recombinant protein purification	Polystyrene-divinylbenzene Methacrylate	<i>p</i> -Aminobenzamidine (pABA)
Styros-AL, -EP, and -OH	OraChrom	Activated Affinity	Protein	Polystyrene-divinylbenzene Methacrylate	Aldehyde, epoxy and hydroxy, respectively
Venusil HILIC	Agela Technologies	HILIC	Polar compounds	Silica	Amide
Viva PFP Propyl	Restek	Wide pore	Large molecules, basic, halogenated compounds	Silica	Pentafluorophenyl
Zirchrom Chiral (S) LEU	Zirchrom Separations	Chiral	Enantiomer separation	Zirconia	(S)-3,5-nitrobenzoyl-leucine
Zirchrom Chiral (S) or (R) NESAA	Zirchrom Separations	Chiral	Enantiomer separation	Zirconia	See description
Zirchrom Chiral (S) or (R) PG	Zirchrom Separations	Chiral	Enantiomer separation	Zirconia	See description

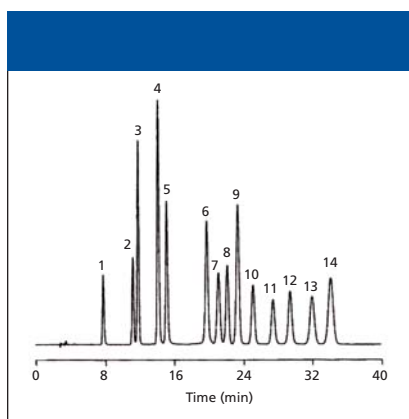


Figure 3: Separation of explosives in EPA Method 8330 using a Dionex Acclaim Explosives column (5- μ m d_p). Column dimensions: 250 mm \times 4.6 mm; mobile phase: 50:50 methanol-water; flow rate: 1 mL/min; temperature: 30 $^{\circ}$ C; injection volume: 5 μ L; detection: UV absorbance at 254 nm. Peaks (50 ppm each): 1 = HMX, 2 = RDX, 3 = 1,3,5-trinitrobenzene, 4 = 1,3-dinitrobenzene, 5 = nitrobenzene, 6 = 2,4,6-trinitrobenzene, 7 = tetryl, 8 = 2,6-dinitrotoluene, 9 = 2,4-dinitrotoluene, 10 = 2-nitrotoluene, 11 = 4-nitrotoluene, 12 = 3-nitrotoluene, 13 = 4-amino-2,6-dinitrotoluene, 14 = 2-amino-4,6-dinitrotoluene. (Courtesy of Dionex Corporation.)

2006 (see Table II). If one also adds all the reversed-phase columns covered in the new series (Table I), in specialty columns (Table III) and in the preparative chromatography section, one can clearly see the dominance of this operational mode. Silica-based bonded phase columns will dominate the introductions again this year but a few hybrid, zirconia and polymeric columns also will be on the scene.

The most popular reversed-phase bonded phase always has been C18 (octadecylsilane) and this year proved it to be true once again with 18 C18 columns being introduced in all categories. However, this phase is not always the optimum for all separations and C8, polar-embedded C18, and cyanopropyl phases will be made available to present different selectivity alternatives. In general, C8 has similar selectivity to the C18 phase but somewhat lower retention at the same mobile phase composition. The polar-embedded phases are particularly suited for the separation of basic compounds. These phases are useful for the

separation of polar compounds where a low percentage of organic modifier is required and where phase collapse (dewetting) can be a problem. The CN phases offer a short-chain reversed-phase packing that has some polar character by virtue of the cyano functionality. Besides lower retention, selectivity can be quite different when compared with C8 and C18 phases. The cyano column can also be used for normal-phase chromatographic separations by careful changeover from the aqueous-based mobile phase to entirely organic mobile phase.

Over the past several years, a number of longer-chain alkyl phases as large as C30 have been introduced. These long-chain phases provide a greater degree of shape selectivity than a typical C18 phase, have long proven superior for the separation of *cis-trans* carotenoid isomers, are resistant to phase collapse at low percentage of organic solvent in the mobile phase and have high loadability. This year another C30 phase joins the ranks.

Most chromatographers facing a separation problem almost immediately go to

d_p (μm)	Pore Size (\AA)	Comments
Not applicable	Gigaporous	Simulated monolith recommended for removal or purification of serine protease such as thrombin after tag cleavage of recombinant proteins; covalent bonding; hard gel offers high mechanical and chemical stability; high loading capacity; column dimensions: 1–250 mm \times 0.01–20 mm; cartridge design; PEEK and stainless steel construction; BIO.
Not applicable	Gigaporous	Simulated monolith materials with the three different active functional groups can immobilize a wide range of ligands via covalent linkage using simple coupling procedures; gigaporous hard gels offer high mechanical and chemical stability; high loading capacity; column dimensions: 1–250 mm \times 0.01–20 mm; cartridge design; PEEK and stainless steel construction; BIO.
5	100	Recommended for use in reversed- or normal-phase modes; 100% water compatible; retentivity for very hydrophilic compounds; could be useful for improved replacement of ion-pair methods, especially for LC-MS applications in HILIC mode; capillary to preparative column dimensions; guard columns available.
3,5	300	Fluorinated phase has propyl spacer; highly retentive for basic analytes and halogenated compounds; wide pores suitable for large molecules (such as proteins); microbore to preparative dimensions (30–300 mm \times 1–50 mm); guard columns and bulk packing available; BIO.
3,5	300	Chiral phase anchored at Lewis-acid site on zirconia base material; phase can be removed by alkaline treatment and can be regenerated; different selectivity than other materials; can be used in LC, SFC, and CE-CEC; column dimensions: and 100–250 mm \times 0.3–4.6 mm.
3,5	300	Functional group: (S)- or (R)-N-[1-(1-naphthyl)ethyl]succinamic acid; chiral phase anchored at Lewis-acid site on zirconia base material; phase can be removed by alkaline treatment and can be regenerated; different selectivity than other materials; can be used in LC, SFC, and CE-CEC; column dimensions: 100–250 mm \times 0.3–4.6 mm.
3,5	300	Functional group: (S)- or (R)-3,5-dinitrobenzoyl-phenylglycine; chiral phase anchored at Lewis-acid site on zirconia base material; phase can be removed by alkaline treatment and can be regenerated; different selectivity than other materials; can be used in LC, SFC, and CE-CEC; column dimensions: 100–250 mm \times 0.3–4.6 mm.

a C18 stationary phase. Sometimes, though, it might be necessary to go beyond the standard column to achieve better selectivity. Figure 1 shows an example of the separation of corticosteroids on a C18 phase, a phenyl phase, and a new biphenyl phase from Restek (Bellefonte, Pennsylvania). The biphenyl functionality appears to offer a slightly different selectivity than the C18 and a quite different selectivity to the phenyl. The Allure Biphenyl phase appears to provide a unique π - π selectivity for unsaturated compounds. For the corticosteroids, the column did offer a better separation under the reversed-phase conditions employed. Although the structures are not shown, these compounds are very similar and were separated in a simple isocratic analysis.

Temperature has gotten recent attention as a chromatographic variable in method development. When the temperature is increased, solvent viscosity is decreased resulting in a lower column back pressure, solute retention is reduced, and column efficiency is

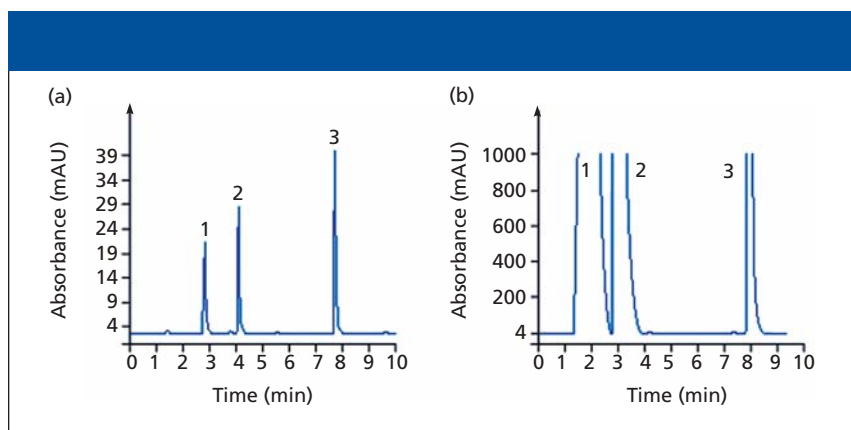


Figure 4: Scale-up of drug separation from (a) an analytical-scale column to (b) a Chromolith SemiPrep monolith column. Column: (a) 100 mm \times 4.6 mm Performance, (b) 100 mm \times 10 mm SemiPrep; mobile phase A: acetonitrile with 0.1% TFA; mobile phase B: water with 0.1% TFA; gradient: 15–80% A in 10 min; flow rate: (a) 1 mL/min, (b) 4.7 mL/min; injection volume: (a) 2 μL , (b) 100 μL ; detection: UV absorbance at 270 nm. Peaks: 1 = nadolol (1 mg/mL in [a], 100 mg/mL in [b]), 2 = metoprolol (1 mg/mL in [a], 100 mg/mL in [b]), 3 = propranolol (0.5 mg/mL in [a], 50 mg/mL in [b]). (Courtesy of Merck KGaA.)

improved. On the downside, higher temperatures can affect solute stability, a fear of some researchers. In addition, many, but not all, silica-based packings and phases have life problems, especially at higher pH values. Polymeric and other

high-temperature-stable columns have efficiency or surface chemistry problems and most HPLC column ovens—heaters have not had very high temperature capability. The situation has changed with the introduction of temperature-sta-

ble columns and new oven configurations capable of high temperature operation. Restek (Bellefonte, Pennsylvania) has come up with a high temperature reversed-phase material that will withstand temperatures as high as 200 °C over a wide pH range.

Ion-Exchange Chromatography—Ion Chromatography

Besides ion-exchange specialty columns in Tables I and III, Dionex (Sunnyvale, California) and Sepax Technologies will introduce new ion-exchange columns. The Dionex IonPac stationary phases are prepared using a unique polymer bonding technology. In their preparation, a novel hyperbranched anion-exchange polymer is electrostatically attached to a surface-sulfonated wide-pore polymeric substrate. This new polymer bonding technology allows control the column selectivity and capacity through alternating treatments of epoxy monomer and amine. Both columns are anion exchangers with alkanol quaternary ammonium functionality and ion-exchange capacities of 210 μ equiv (AS-22) and 315 μ equiv (AS-23). Both 2-mm and 4-mm i.d. columns were included as well as matching guard columns. The columns are suited for the separation of common inorganic anions in drinking water and other matrices according to EPA Method 300.0 using simple carbonate–bicarbonate buffers and suppressed conductivity detection. The carbonate peak is well resolved from the other ions. The AS-23 column is further recommended for those samples also containing oxyhalides such as chlorite, bromate, and chlorate according to EPA Method 300.1. The columns have pH stabilities over the entire pH range and therefore are suitable for very caustic and acidic samples.

The Sepax Proteomix ion-exchange offerings are based upon very highly crosslinked (80%) nonporous polystyrene–divinylbenzene (PS-DVB) spherical resins with particle diameters of 1.0, 1.7, and 3- μ m. The smaller sizes are recommended for the ultrahigh-pressure and high-speed LC of proteins, peptides, nucleic acids, and carbohydrates. Four chemistries, two cation exchangers (sulfonic strong cation and carboxyl weak cation), and two anion exchangers (qua-

ternary ammonium strong anion and diethylamine weak anion) are available. The columns will be available with nano- (50–75- μ m), capillary (0.15–0.75 mm), microbore (1.0–2.1 mm), and conventional (4.6 mm) internal diameters and 3–10 mm lengths with stainless steel or PEEK construction. The proprietary surface chemistry on the nonporous resins enables separation capacity comparable to that of porous resins.

Normal-Phase–Adsorption Chromatography

In addition to bare silica gel columns introduced in Table I, there will be one notable introduction by J.T. Baker/Mallinkrodt, the company's BakerBond Spherical Flash silica. Sold in bulk and in prepacked cartridges, the spherical packing material should pack more reproducibly and provide higher mechanical strength and fewer fines, thereby allowing higher flow rates at lower back pressure than the more typical irregular silica gel materials. The silica gel has a 50- μ m particle size and 60-Å pores.

Size Exclusion Chromatography

Each year only a few new SEC columns are introduced at Pittcon. This year, a total of six phases will be introduced, four by Sepax Technologies (Newark, Delaware) and two by Polymer Laboratories (Amherst, Massachusetts), now part of Varian (Palo Alto, California). The Sepax offerings, called the CNT line, are really specialty columns for the SEC separation of carbon nanotubes, nanoparticles, and nanorods based upon their lengths. The recovery for the length-dependent separation of DNA-wrapped carbon nanotubes is nearly 100%. The columns are packed with spherical silica particles of 5-, 7-, or 10- μ m diameter with pore sizes of 300, 500, 1000, and 2000 Å. The particles have a proprietary bonded stationary phase. Stainless steel columns are available in a variety of internal diameters of 4.6–21.2 mm. Another carbon nanotube separation column based upon ion-exchange properties (rather than SEC) can be found in the Specialty columns section (Table III).

Polymer Labs will introduce one nonaqueous PS-DVB PLgel SEC column that is devoted exclusively to the separation of polyolefins in trichlorobenzene.

The 13- μ m material is suited for polymers as large as 10⁹ g/mol. The 300 mm \times 7.5 mm column is designed to provide improved resolution and an increased operating range over previously offered columns of this type. A guard column packed with the same material is also provided.

Polymer Labs second offering is a proprietary macroporous polyhydroxyl column for use in aqueous SEC. The column's 5- μ m particles, small for a typical SEC column, equate to a higher resolution of lower molecular weight water-soluble polymers as large as 20,000 g/mol. The column also has the 300 mm \times 7.5 mm dimensions.

Specialty HPLC Columns

Specialty columns are HPLC columns that have been developed for specific separations that are difficult to achieve on a standard column. However, sometimes manufacturers will use a standard column but test it specifically for a certain class or compounds and provide a recommended set of chromatographic conditions. In some cases, the specialty column comes as part of a "total solution" kit with reagents, standards, and method. Most specialty columns are delivered with a test chromatogram from an analysis performed at the factory before shipment, and some are guaranteed. Table III shows the 26 specialty columns that will be presented at Pittcon 2006. Silica gel–based columns will dominate, but polymer-based, hybrid, and zirconia specialty columns also will be introduced.

The largest categories of specialty columns are those designated for the separation of biomolecules (one-third of total), mainly proteins and peptides, and those for the separations of chiral compounds (one-third of total).

A variety of selective columns for protein analysis and purification using principles such as affinity and immobilized metal ion affinity chromatography (IMAC) will be introduced at Pittcon 2006. Some representative examples include columns introduced by Dionex Corp (Sunnyvale, California), who displayed an IMAC column specific for His-tagged proteins and a second mixed mode monolith column that can be used at high flow rates with a low pressure drop, Orachrom's (Woburn, Massachu-

sets) simulated monolith gigaporous columns for protein purifications by affinity-derivatization and tagged proteins, and, and Agilent's mRP-C18 (Santa Clara, California) column for membrane and other sticky proteins. Waters' (Milford, Massachusetts) peptide mapping column is based upon their silica-hybrid particle BEH technology.

A larger than normal number of chiral columns are slated for introduction at Pittcon 2006. The regular chiral companies such as Chiral Technologies (West Chester, Pennsylvania) and Astec (Whippany, New Jersey) will be present but this year Zirchrom (Anoka, Minnesota) joined their ranks with zirconia-based chiral columns. Zirchrom uses the strong Lewis acid sites on the zirconia base material to their advantage by using that group to anchor the chiral selector. Interestingly, the chiral phase can be removed by alkaline treatment to regenerate a contaminated or spent column or to convert the column into another chiral phase by rinsing with a chiral selector solution. Chiral Technologies has expanded their popular Chiralpak phases to include capillary dimensions. Because supercritical fluid chromatography (SFC) is a popular technique for chiral purifications, especially in the preparative mode, several of the chiral columns can be used for SFC separations in addition to HPLC separations.

To illustrate the advantages of a specialty chiral column, the right-hand chromatogram in Figure 2c shows the chiral separation of *trans*-stilbene oxide on a Cyclobond I 2000 DNP column. There was no separation on the Cyclobond I 2000 column, which is a β -cyclodextrin column. However, by the derivatization of the β -cyclodextrin to provide dinitrophenyl functionality bonded through an ether linkage (Figure 2a), the two enantiomers of *trans*-stilbene oxide were well resolved using an isocratic mobile phase system. With this derivatized phase, a π -sharing system is established with analytes having π -systems in the stereogenic environment. *trans*-Stilbene oxide with its aromatic rings satisfies this requirement. This phase demonstrates enhanced selectivity over the conventional β -cyclodextrin phase.

This year, a number of HILIC columns will be introduced. HILIC is a

separation technique for highly polar analytes that get around some of the problems associated with reversed-phase chromatography such as low retention or phase collapse (dewetting). HILIC uses a polar stationary phase such as bare silica gel or polar bonded phase (for example, diol) and requires a high percentage of a nonpolar mobile phase, similar to the requirements for normal-phase chromatography. However, unlike normal-phase chromatography, which uses nonpolar solvents like hexane and methylene chloride and tries to exclude water from the mobile phase, HILIC requires some water in the mobile phase to maintain a stagnant enriched water layer on the packing surface into which analytes might selectively partition. In addition, water-miscible organic solvents are used. Under HILIC, polar analytes are well retained and are eluted in order of increasing hydrophilicity. ES Industries (West Berlin, New Jersey) and Angela Technologies (Newark, Delaware) plan to introduce columns for HILIC with amide and hydroxyl functionality. In a slight twist, MicroSolv Technology (Eatontown, New Jersey) will introduce the Cogent UDA column for a technique called "nonaqueous normal phase" where the packing is terminated with hydride group (Si-H) rather than silanols.

Specialty columns for explosives, LC-MS, carbon nanotubes (by virtue of their interaction with oligonucleotides on the DNA wrapping), oligonucleotides and halogenated compounds round out the offerings. The Dionex Acclaim Explosives column provides a baseline separation of the 14 explosive compounds outlined in EPA Method 8330 "Nitroaromatics and Nitramines by HPLC." The separation on the silica-based Acclaim column is shown in Figure 3. The method is useful for the separation of explosives in these explosive compounds in surface and ground water.

Preparative Chromatography

Four new column products and one column packing device, specifically for preparative chromatography were introduced at this year's Pittcon. Merck KGaA (Darmstadt, Germany) expanded their line of Chromolith silica monolithic columns to include a semipreparative

reversed-phase version. Previously, the company introduced a capillary version (0.1-mm i.d.), in analytical version (4.6-mm i.d.) and preparative version (25-mm i.d.). The Chromolith Semi-Prep RP-18 has the dimensions of 10-mm i.d. by 100-mm length. For the best peak shape for basic analytes, the silica monolith is endcapped. The column is clad with PEEK polymer. The monolith has a surface area of 300 m²/g and 18% carbon coverage (3.6- μ m/m²). Although the column pressure rating is 150 bar, due to its monolithic structure operating pressure is generally much lower. Figure 4 shows the scaleup of a pharmaceutical separation from a 4.6-mm i.d. analytical column to the 10-mm i.d. semipreparative column of the same length. The injection of 10 mg of each compound (30 mg total) is shown in the figure; up to 80 mg per compound has been reported for this semipreparative column in less than 30 min (3).

Waters (Milford, Massachusetts) has expanded the SunFire Prep column line and has added a silica gel column. In preparative LC, adsorption chromatography is a popular mode. The mobile phase solvents that are used are usually volatile and therefore more easily removed from collected fractions than when the compounds are separated by reversed-phase HPLC with aqueous solvents. The Waters column is based on high purity (Type B) silica of particle sizes of 5- and 10- μ m. The pore size is 100 Å, ideal for small molecules. The columns are packed to their optimum bed density (OBD) and a scaleable such that separations achieved on other SunFire columns can be matched on the preparative version. Various dimensions are available.

Phenomenex (Torrance, California) added a SecurityGuard Prep guard column. Since samples injected into preparative columns often contain impurities and other compounds that may foul the expensive preparative column, a guard column is always advisable to protect the preparative column against contamination. Phenomenex's guard column is provided in a 21.2-mm i.d. \times 15-mm length for all their common preparative phases available in 18–50-mm i.d.'s. The cartridge design makes it convenient to change guard columns.

Phenomenex also displayed the AXIA column packing and hardware system that uses a hydraulic piston to keep pressure on the packed bed. Often preparative columns operated continuously at high flow rate conditions can have fundamental problems such as collapse, voiding, and channeling of the packed bed resulting in variable or lower efficiency. With this computer-controlled new system, the bed density is custom calculated and adjusted for column size. With controlled hydraulic compression, the support media is never allowed to decompress resulting in overall column-to-column consistency.

Acknowledgment

I would like to thank the manufacturers and distributors that kindly furnished the requested information in advance of Pittcon 2006, thus, allowing a timely report on new product introductions. For those manufacturers who would like to be considered for inclusion into Pittcon 2007 coverage, please send the name of the primary company contact, the mailing address, FAX and e-mail numbers to Patrick Kempf, Managing Editor, LCGC North America, c/o Advanstar Communications, 485 Rte. 1 South, Bldg. F, Iselin, New Jersey 08830, Attn.: Pittcon '07 "ColumnWatch."

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