

# HICHROM

**Chromatography Columns and Supplies** 





# **Hichrom Limited**

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- Spherical porous silica
- Narrow particle and pore size distribution
- · Hichrom high efficiency

ZORBAX® spherical particles are manufactured from small uniform colloidal silica microbeads which are agglutinated in a Du Pont patented organic polymerisation process. The organic polymer is removed by a controlled sintering process to yield mechanically stable silica particles. ZORBAX is manufactured by Agilent Technologies Inc. Their original ZORBAX phases (listed below) are based on Type A silica.

#### **ZORBAX Phases**

ZORBAX Phase	Functional Group	Endcapped	Particle Size (µm)	Pore Size (Å)	Surface Area (m²/g)	Carbon Load (%)
Silica	-	-	5	70	330	-
TMS	Methyl	Yes	5	70	330	4
C8	Octyl	Yes	5	70	330	12
ODS	Octadecyl	Yes	5	70	330	20
Phenyl	Phenyl	Yes	5	70	330	12
NH2	Amino	Yes	5	70	330	4
CN	Cyano	Yes	5	70	330	7

# Ordering Information - Hichrom Manufactured Columns

#### Microbore (1.0mm i.d.) Columns

Please contact Hichrom for further details of 1.0mm i.d. ZORBAX columns.

#### Microbore (2.1mm i.d.) Columns

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Fum 70DDAY Dhoop		Column Dime	nsions¹ (mm)		Guard Cartridges <sup>2</sup>
5μm ZORBAX Phase	50 x 2.1	100 x 2.1	150 x 2.1	250 x 2.1	(For 2.1mm i.d. Columns)
					(5/pk) <sup>3</sup>
Silica	ZSIL-50AM	ZSIL-100AM	ZSIL-150AM	ZSIL-250AM	ZSIL-10CM5
TMS	ZTMS-50AM	ZTMS-100AM	ZTMS-150AM	ZTMS-250AM	ZTMS-10CM5
C8	ZC8-50AM	ZC8-100AM	ZC8-150AM	ZC8-250AM	ZC8-10CM5
ODS	ZODS-50AM	ZODS-100AM	ZODS-150AM	ZODS-250AM	ZODS-10CM5
Phenyl	ZPH-50AM	ZPH-100AM	ZPH-150AM	ZPH-250AM	ZPH-10CM5
NH2	ZNH-50AM	ZNH-100AM	ZNH-150AM	ZNH-250AM	ZNH-10CM5
CN	ZCN-50AM	ZCN-100AM	ZCN-150AM	ZCN-250AM	ZCN-10CM5

#### Medium Bore (3.2mm i.d.) Columns

Fum 70DDAY Dhoop		Column Dime	ensions¹ (mm)		Guard Cartridges <sup>2</sup>
5μm ZORBAX Phase	50 x 3.2	100 x 3.2	150 x 3.2	250 x 3.2	(For 3.2mm i.d. Columns)
					(5/pk) <sup>3</sup>
Silica	ZSIL-50AS	ZSIL-100AS	ZSIL-150AS	ZSIL-250AS	ZSIL-10C5
TMS	ZTMS-50AS	ZTMS-100AS	ZTMS-150AS	ZTMS-250AS	ZTMS-10C5
C8	ZC8-50AS	ZC8-100AS	ZC8-150AS	ZC8-250AS	ZC8-10C5
ODS	ZODS-50AS	ZODS-100AS	ZODS-150AS	ZODS-250AS	ZODS-10C5
Phenyl	ZPH-50AS	ZPH-100AS	ZPH-150AS	ZPH-250AS	ZPH-10C5
NH2	ZNH-50AS	ZNH-100AS	ZNH-150AS	ZNH-250AS	ZNH-10C5
CN	ZCN-50AS	ZCN-100AS	ZCN-150AS	ZCN-250AS	ZCN-10C5

<sup>&</sup>lt;sup>1</sup> Other column dimensions available – please enquire

#### Analytical (4.0mm i.d.) Columns

Please contact Hichrom for further details of 4.0mm i.d. ZORBAX columns.

<sup>&</sup>lt;sup>2</sup> Use with free-standing holder HI-161 and column coupler HI-081 - see p.24

<sup>3</sup> Starter kits also available - see p.25

# **ZORBAX®** – Hichrom Manufactured Columns (continued)

# Analytical (4.6mm i.d.) Columns

5μm ZORBAX Phase		Column Dimensions¹ (mm)					
	50 x 4.6	100 x 4.6	150 x 4.6	250 x 4.6	(For 4.6mm i.d. Columns)		
					(5/pk) <sup>3</sup>		
Silica	ZSIL-50A	ZSIL-100A	ZSIL-150A	ZSIL-250A	ZSIL-10C5		
TMS	ZTMS-50A	ZTMS-100A	ZTMS-150A	ZTMS-250A	ZTMS-10C5		
C8	ZC8-50A	ZC8-100A	ZC8-150A	ZC8-250A	ZC8-10C5		
ODS	ZODS-50A	ZODS-100A	ZODS-150A	ZODS-250A	ZODS-10C5		
Phenyl	ZPH-50A	ZPH-100A	ZPH-150A	ZPH-250A	ZPH-10C5		
NH2	ZNH-50A	ZNH-100A	ZNH-150A	ZNH-250A	ZNH-10C5		
CN	ZCN-50A	ZCN-100A	ZCN-150A	ZCN-250A	ZCN-10C5		

#### Semi-Preparative (7.75mm i.d.) Columns

5μm ZORBAX Phase		Column Dime	ensions¹ (mm)		Guard Cartridges <sup>4</sup>
	50 x 7.75	100 x 7.75	150 x 7.75	250 x 7.75	(For 7.75mm i.d. Columns)
					(3/pk)
Silica	ZSIL-50SP	ZSIL-100SP	ZSIL-150SP	ZSIL-250SP	ZSIL-10CP3
TMS	ZTMS-50SP	ZTMS-100SP	ZTMS-150SP	ZTMS-250SP	ZTMS-10CP3
C8	ZC8-50SP	ZC8-100SP	ZC8-150SP	ZC8-250SP	ZC8-10CP3
ODS	ZODS-50SP	ZODS-100SP	ZODS-150SP	ZODS-250SP	ZODS-10CP3
Phenyl	ZPH-50SP	ZPH-100SP	ZPH-150SP	ZPH-250SP	ZPH-10CP3
NH2	ZNH-50SP	ZNH-100SP	ZNH-150SP	ZNH-250SP	ZNH-10CP3
CN	ZCN-50SP	ZCN-100SP	ZCN-150SP	ZCN-250SP	ZCN-10CP3

#### Semi-Preparative (10.0mm i.d.) Columns

Fum 70DDAY Dhoop		Column Dime	ensions¹ (mm)		Guard Cartridges <sup>4</sup>
5μm ZORBAX Phase	50 x 10.0	100 x 10.0	150 x 10.0	250 x 10.0	(For 10.0mm i.d. Columns)
					(3/pk)
Silica	ZSIL-50SP1	ZSIL-100SP1	ZSIL-150SP1	ZSIL-250SP1	ZSIL-10CP3
TMS	ZTMS-50SP1	ZTMS-100SP1	ZTMS-150SP1	ZTMS-250SP1	ZTMS-10CP3
C8	ZC8-50SP1	ZC8-100SP1	ZC8-150SP1	ZC8-250SP1	ZC8-10CP3
ODS	ZODS-50SP1	ZODS-100SP1	Z0DS-150SP1	Z0DS-250SP1	ZODS-10CP3
Phenyl	ZPH-50SP1	ZPH-100SP1	ZPH-150SP1	ZPH-250SP1	ZPH-10CP3
NH2	ZNH-50SP1	ZNH-100SP1	ZNH-150SP1	ZNH-250SP1	ZNH-10CP3
CN	ZCN-50SP1	ZCN-100SP1	ZCN-150SP1	ZCN-250SP1	ZCN-10CP3

# Preparative (21.2mm i.d.) Columns

5μm ZORBAX Phase		Column Dime	ensions¹ (mm)		Guard Cartridges <sup>4</sup>
	50 x 21.2	100 x 21.2	150 x 21.2	250 x 21.2	(For 21.2mm i.d. Columns)
					(3/pk)
Silica	ZSIL-50P	ZSIL-100P	ZSIL-150P	ZSIL-250P	ZSIL-10CP3
TMS	ZTMS-50P	ZTMS-100P	ZTMS-150P	ZTMS-250P	ZTMS-10CP3
C8	ZC8-50P	ZC8-100P	ZC8-150P	ZC8-250P	ZC8-10CP3
ODS	ZODS-50P	ZODS-100P	ZODS-150P	ZODS-250P	ZODS-10CP3
Phenyl	ZPH-50P	ZPH-100P	ZPH-150P	ZPH-250P	ZPH-10CP3
NH2	ZNH-50P	ZNH-100P	ZNH-150P	ZNH-250P	ZNH-10CP3
CN	ZCN-50P	ZCN-100P	ZCN-150P	ZCN-250P	ZCN-10CP3

<sup>&</sup>lt;sup>1</sup> Other column dimensions available – please enquire

<sup>&</sup>lt;sup>2</sup> Use with free-standing holder HI-161 and column coupler HI-081 — see p

<sup>&</sup>lt;sup>3</sup> Starter kits also available – see p.25

<sup>&</sup>lt;sup>4</sup> Use with free-standing holder HI-150 and column coupler HI-081

# **ZORBAX® – Agilent Manufactured Columns**

Agilent Technologies manufacture several ranges of silica based columns for analytical and preparative HPLC. A range of traditional phase ZORBAX® columns are also available from Hichrom (please see pages 279-280).

The following table summarises the features of various Agilent ZORBAX and Poroshell column ranges for reversed-phase HPLC. Please contact us for further technical details on any ZORBAX or Poroshell column.

#### **Reversed-Phase HPLC Series**

Column Range	Particle Type	Endcapping	Side Group Structure	Phases	Pore Size (Å)	Surface Area (m²/g)
Eclipse Plus	Totally porous	Double <sup>1</sup>	Dimethyl	C18, C8, PAH, Phenyl-Hexyl	95	160
Eclipse XDB	Totally porous	Double	Dimethyl	C18, C8, Phenyl, CN	80	180
StableBond (SB)	Totally paraua	None	Diisopropyl (C8, C3, CN,	C18, C8, C3, Phenyl, CN, Aq	80	180
300StableBond (300SB)	Totally porous	None	phenyl, Aq), diisobutyl (C18)	C18, C8, C3, CN	300	45
Bonus-RP	Totally porous	Triple	Diisopropyl	proprietary	80	180
Extend-C18	Totally porous	Double	Unique bidentate structure	C18	80	180
300Extend-C18	Totally porous	Double	Unique bidentate structure	C18	300	45
Rx	Totally porous	None	Dimethyl	C18, C8	80	180
Poroshell 120 EC	Superficially porous	Double	Dimethyl	C18	120	120
Poroshell 120 SB	Superficially porous	None	Diisobutyl	C18, C8	120	120
Poroshell 300	Superficially porous	As for 3	00SB or 300Extend-C18	300SB-C18, C8, C3, 300Extend-C18	300	45

<sup>1</sup> Except PAH phase

# **Overview of Reversed-Phase Bonded Phases**

### **ZORBAX Eclipse Plus**

- · Improved silica and bonding technology
- Extra dense bonding and double endcapping
- Superior peak shape for basic compounds
- Long life from pH 2 9
- High resolution and efficiency with 1.8, 3.5 and 5µm columns
- Rigorous QA/QC testing

# **ZORBAX Eclipse XDB**

- Good peak shape for acids, bases and neutrals
- High performance over a wide pH range (2 9)
- · Long lifetime with extra dense bonding and double endcapping
- Four bonded phases for flexible method development
- Increased column stability at mid pH
- Fast, high resolution separations using 1.8 and 3.5µm columns
- Capillary to preparative dimensions

# **ZORBAX StableBond (SB)**

- $\bullet$  Exceptional stability at low pH (1 2)
- Use of high temperature (up to 90°C for C18, 80°C for C8, C3, Phenyl, CN and Aq)
- · Widest selection of bonded phases for different selectivity
- Uses LC-MS friendly eluents, with formic acid, acetic acid or TFA
- Rapid separations using 1.8 and 3.5µm columns
- Nano to preparative dimensions

#### ZORBAX 300StableBond (300SB)

- Wide-pore (300Å) phases for peptide and protein separations
- All the benefits of ZORBAX StableBond phases
- Robust separations at low pH with TFA-containing eluents
- Capillary and nano dimensions for proteomics separations

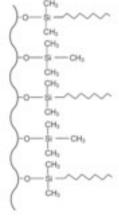


Figure 1. Eclipse Plus and Eclipse XDB bonded phases

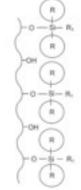


Figure 2. Sterically protected StableBond bonded phase

### **ZORBAX®** – Agilent Manufactured Columns (continued)

#### **ZORBAX Bonus-RP**

- Novel bonding technology with embedded polar amide group in long alkyl chain
- Excellent peak shape for challenging basic compounds at low and mid pH
- . Sterically protected bonding for use at low pH
- Usable in 100% aqueous eluents
- Triple endcapped for good peak shape
- Microbore to preparative dimensions
- 1.8, 3.5 and 5µm particle sizes

ZORBAX® Bonus-RP has a polar amide group embedded in a long alkyl chain. The diisopropyl side groups (see Figure 3) provide steric protection against acid hydrolysis for good lifetime at low pH. The Bonus-RP column provides an alternative selectivity to C18 and C8 alkyl bonded phases.

#### ZORBAX Extend-C18 and 300Extend-C18

- Unique bidentate bonding and double endcapping for high pH stability
- Stable up to pH 11.5
- Ammonium hydroxide recommended as eluent additive with LC-MS of small molecules
- Capillary to preparative dimensions
- Rapid separations using 3.5µm columns
- 80Å and 300Å phases

ZORBAX Extend-C18, by operating at high pH, enables basic compounds to be analysed with good peak shape above their pKa in the free base form. High pH separations are also appropriate for compounds that are more stable or more soluble in high pH solutions. In addition to ammonium hydroxide, triethylamine, pyrrolidine, glycine and borate buffer options can be used.

#### **ZORBAX Rx**

- General separations of basic, acidic and neutral compounds at low pH
- Different selectivity than SB columns
- Rx-C8 is the same as SB-C8
- Analytical to preparative dimensions

### Poroshell 120

- Superficially porous particles for high efficiency at low pressure
- Sub 2µm efficiency with a 2.7µm particle size
- Endcapped C18 (EC-18) and non-endcapped C18 (SB-C18) and C8 (EC-C8)
- Ideal for use up to 600 bar for HPLC and UHPLC

Agilent Poroshell 120 columns are a 2.7µm particle with a 1.7µm solid core and 0.5µm porous outer layer. Efficiencies similar to sub 2µm columns may be achieved, at 40-50% less pressure. Narrow particle size distribution improves resolution and efficiency, while the core limits diffusion distance and improves separation speed.

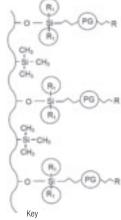
#### Poroshell 300

- 5µm superficially porous particles for separation of biomolecules
- High efficiency and recovery for proteins (up to 1,000kDa) and monoclonal antibodies
- Ideal for LC-MS
- Achieve long lifetime at low pH with Poroshell 300SB and at high pH with 300Extend-C18
- Four different bonded phases 300SB-C18, 300SB-C8, 300SB-C3 and 300Extend-C18

# **ZORBAX PrepHT**

- Easy scale-up from analytical to preparative scale with ZORBAX phases
- Fast preparative separations
- 5 and 7µm particles for high efficiency and high yield
- Variety of bonded phases Eclipse XDB, StableBond, Bonus-RP and Extend-C18

Please contact Hichrom for further technical details on any ZORBAX or Poroshell column.



 $\begin{array}{l} R_1 = \text{Steric protection group} \\ PG = Polar group \\ R = Hydrophobic chain \\ The phase is triple endcapped \end{array}$ 

Figure 3. ZORBAX Bonus-RP structure

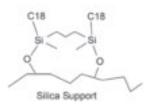
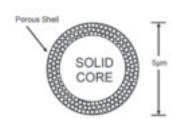


Figure 4. The unique bidentate structure of ZORBAX Extend-C18



# **ZORBAX®** – Agilent Manufactured Columns (continued)

The following table summarises the column hardware descriptions used for different particle size and dimension ZORBAX® columns.

Hardware Description	Column i.d. (mm)	Column Lengths (mm)	Particle Size (µm)	Column Ranges
Semi-preparative	9.4	250, 150, 100, 50	5	Eclipse XDB, StableBond, Rx
Analytical	4.6	250, 150, 100, 50	5	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18, Rx
Rapid Resolution	4.6	250, 150, 100, 75, 50, 30, 20	3.5	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18, Rx
Rapid Resolution HT, 600 bar	4.6	150, 100, 75, 50, 30, 20	1.8	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18
Solvent Saver	3.0	250, 150	5	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18, Rx
Solvent Saver Plus	3.0	150, 100, 75, 50	3.5	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18, Rx
Solvent Saver RRHD, 1200 bar	3.0	150, 100, 50	1.8	Eclipse Plus, Eclipse XDB, StableBond, Extend-C18
Solvent Saver HT, 600 bar	3.0	150, 100, 50, 30, 20	1.8	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18
Narrow Bore	2.1	250, 150, 75, 50	5	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18, Rx, Poroshell 300
Narrow Bore RR	2.1	150, 100, 75, 50, 30, 20	3.5	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18, Rx
Narrow Bore RRHD, 1200 bar	2.1	150, 100, 50	1.8	Eclipse Plus, Eclipse XDB, StableBond, Extend-C18
Narrow Bore RRHT, 600 bar	2.1	150, 100, 50, 30, 20	1.8	Eclipse Plus, Eclipse XDB, StableBond, Bonus-RP, Extend-C18
MicroBore	1.0	250, 75	5	300StableBond, Poroshell 300
MicroBore RR	1.0	150, 50, 30	3.5	Eclipse XDB, StableBond, Bonus-RP, Extend-C18
Capillary	0.3, 0.5	250, 150, 75, 35	5	Eclipse XDB, 300StableBond, Poroshell 300
Capillary RR	0.3, 0.5	150, 100, 75, 50, 35	3.5	Eclipse XDB, 300StableBond, 300Extend-C18
Nano RR	0.1, 0.075	150, 50	3.5	300StableBond

Please note that not all column ranges are available in all possible dimensions.

Please enquire for ordering information for any of these column ranges.

### **ZORBAX GF-250 and GF-450 Gel Filtration columns**

- High efficiency and reproducibility with short analysis time
- Hydrophilic diol bonded silica phase for good protein recovery
- Compatible with organic modifiers (<25%) and denaturants
- $\bullet$  Wide usable pH range (3 8) due to unique zirconia modification
- Narrow pore size and particle size distributions
- ZORBAX GF-250 pore size 150Å, particle size 4μm, surface area 140m²/g, MW range 4,000 400,000 Da
- ZORBAX GF-450 pore size 300Å, particle size 6µm, surface area 50m²/g, MW range 10,000 900,000 Da

Agilent ZORBAX GF-250 and GF-450 size exclusion (gel filtration) columns are used for the size separations of proteins and other biomolecules. Typical applications include separations of protein monomers, dimers and aggregates, desalting, protein molecular weight estimation and separation of modified proteins.

Please enquire for ordering information.