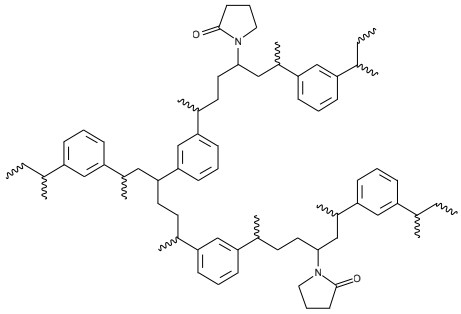
Modern polymeric CHROMABOND® SPE phases

CHROMABOND® HLB

Technical data

Hydrophilic-lipophilic balanced *N*-vinylpyrrolidone-divinylbenzene copolymer (NVP / DVB)

SPE mode:	Reversed phase
Interactions:	Hydrophobic and polar
Particle shape:	Spherical
pH stability:	1–14
Particle size:	60 µm and 30 µm
Pore size:	65 Å
Specific surface:	750 m^2/g



Special characteristics

- Applicable for a wide range of analyte polarities
- High loadability and outstanding performance
- Water wettable – even if bed runs dry, SPE can be continued

Recommended application

- Medium polar organic molecules from polar matrices
- Drugs and pharmaceuticals from urine, blood, serum and plasma
- Tetracyclines and alkaloids from serum
- Pesticides from water

Good to know

A possible replacement for:

- Oasis® HLB
- Strata™-X
- Supel™-Select HLB
- Supra-Poly® HLB
- Isolute® ENV+

Standard SPE procedure for CHROMABOND® HLB (subsequent HPLC analysis)

MN Appl. No. 306300

Column type:
CHROMABOND® HLB / 3 mL / 200 mg, REF 730924

Sample pretreatment:

Individual sample preparation in reference to the compounds and matrix. (Adjust pH value if necessary)

Conditioning:	5 mL methanol, then 5 mL dist. water
Sample application:	Slowly aspirate sample through column
Washing:	5 mL dist. water
Drying:	10 min with applied vacuum
Elution:	8 mL methanol
Evaporation:	Under nitrogen
Reconstitution:	In 1 mL dist. water + 0.1 % formic acid

Standard SPE procedure for CHROMABOND® HLB (subsequent GC analysis)

MN Appl. No. 306310

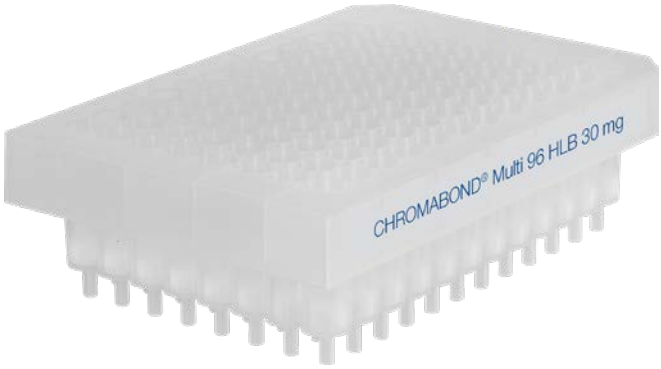
Column type:
CHROMABOND® HLB / 3 mL / 200 mg, REF 730924

Sample pretreatment:

Individual sample preparation in reference to the compounds and matrix. (Adjust pH value if necessary)

Conditioning:	5 mL solvent (e.g., ethyl acetate), 5 mL methanol, 5 mL dist. water
Sample application:	Slowly aspirate sample through column
Washing:	5 mL dist. water
Drying:	10 min with applied vacuum
Elution:	Solvent[1] (typical solvents: ethyl acetate, MTBE, methylene chloride)
Evaporation:	Under nitrogen, dry with sodium sulfate[2], adjust to final volume

[1] usually nonpolar, therefore often 10 % methanol are added
[2] e.g., with CHROMAFIX® Dry



Modern polymeric CHROMABOND® SPE phases

Applications

Tetracyclines and alkaloids from serum at pH 5

MN Appl. No. 306380

Chromatographic conditions

Columns: CHROMABOND® HLB / 1 mL / 30 mg
Oasis® HLB / 1 mL / 30 mg

MN REF: 730921

Conditioning: 1 mL methanol, then 1 mL dist. water

Application: 1 mL serum pH 5, adjusted with formic acid (spiked with 20 µg/mL of each analyte)

Washing: 1 mL dist. water

Drying: 10 min with applied vacuum

Elution: 2 mL methanol

Evaporation: Under nitrogen, 40 °C

Reconstitution: In 1 mL dist. water + 0.1 % formic acid

Recovery rates ± RSD [%], n = 4

Compound	CHROMABOND® HLB	Oasis® HLB
Berberine	85.4 ± 0.3	82.5 ± 0.6
Chlortetracycline	72.1 ± 1.4	66.3 ± 2.8
Hydrastine	88.9 ± 2.6	99.3 ± 5.7
Oxytetracycline	82.3 ± 1.4	78.7 ± 1.4
Tetracycline	78.1 ± 1.4	70.7 ± 2.6

Further analysis: HPLC, according to MN Appl. No. 128180

Column: EC 50/2 NUCLEOSHELL® RP 18plus, 2.7 µm

MN REF: 763232.20

Eluent: A: dist. water + 0.1 % formic acid
B: acetonitrile + 0.1 % formic acid
Gradient: 2–60 % B in 4 min, 60 % B for 1 min, 60–2 % B in 0.5 min, 2 % B for 3 min

Flowrate: 0.75 mL/min

Temperature: 22 °C

Detection: UV, 330 nm

Injection: 5 µL

Mycotoxins in wheat flour

MN Appl. No. 306740

Chromatographic conditions

Columns: CHROMABOND® HLB / 60 µm / 3 mL / 200 mg

MN REF: 730924

Extraction:

- Weigh 4 g homogenized sample in an empty 50 mL centrifuge tube
- Add 8 µL mycotoxin standard mixture (β = 10 µg/mL each analyte in acetonitrile)
- Add 10 mL of water / acetonitrile mixture (20:80, v/v), shake vigorously and wait 10 min
- Add CHROMABOND® QuEChERS extraction Mix XII (REF 730648), shake vigorously for 1 min and cool the mixture down in an ice bath
- Centrifuge at 4500 rpm for 20 min at 20 °C
- Take organic phase for clean-up procedure

Conditioning: 6 mL acetonitrile

Application: 1 mL sample extract was aspirated with low vacuum into a vial

Elution: 4 mL acetonitrile were aspirated with low vacuum into a vial

Evaporation: Combine cleaned sample extract and acetonitrile eluate and evaporate to dryness under nitrogen, 60 °C

Reconstitution: In 1 mL acetonitrile

Analyte	Recovery rate [%]	RSD [%], n = 5
Aflatoxin B1	88	2.6
Aflatoxin B2	91	5.0
Aflatoxin G1	85	2.6
Aflatoxin G2	88	4.5
HT-2 toxin	115	5.7
T-2 toxin	106	5.1
Zearalenone	49	3.4

Modern polymeric CHROMABOND® SPE phases

Applications

Sulfa drugs from serum

MN Appl. No. 306340

Columns*:	CHROMABOND® HLB / 60 µm / 1 mL / 30 mg Oasis® HLB / 60 µm / 1 mL / 30 mg
MN REF:	730921
Conditioning:	1 mL methanol, 1 mL dist. water
Application:	1 mL serum (spiked with 10 µg/mL of each analyte)
Washing:	1 mL dist. water
Drying:	10 min with applied vacuum
Elution:	2 mL methanol
Evaporation:	Under nitrogen, 40 °C
Reconstitution:	In 1 mL dist. water + 0.1 % formic acid

Equivalence to Oasis® HLB

CHROMABOND® HLB shows equivalent recovery rates to Oasis® HLB for the three tested sulfa drugs.

Further analysis: HPLC, according to MN Appl. No. 128130

Column:	EC 150/2 NUCLEODUR® C18 Pyramid, 3 µm
MN REF:	760261.20
Eluent:	Dist. water + 0.1 % formic acid / methanol + 0.1 % formic acid (85:15, v/v), 5 min
Flow rate:	0.6 mL/min
Temperature:	25 °C
Detection	UV, 254 nm
Injection:	5 µL

Recovery rates ± RSD [%], n = 5

Compound	CHROMABOND® HLB	Oasis® HLB
Sulfadiazine	97.3 ± 2.9	92.0 ± 3.8
Sulfamerazine	94.4 ± 1.8	92.8 ± 1.6
Sulfathiazole	90.3 ± 2.9	89.6 ± 1.5

Chloramphenicol from honey

MN Appl. No. 306350

Columns*:	CHROMABOND® HLB / 60 µm / 3 mL, 200 mg Oasis® HLB, 3 mL, 200 mg
MN REF:	730924

Sample pretreatment:

Weigh out 5 g of honey. Add 4 mL water and shake rigorously for 30 sec. Spike with 1 mL standard solution (c = 5 ng/mL in methanol) and shake rigorously for 30 sec. Add 15 mL ethyl acetate and shake rigorously for 30 sec. Centrifuge at 3000 rpm for 10 min. Take 12 mL of supernantant for eluent exchange. Evaporate extracts to dryness at 40 °C under a stream of nitrogen. Redissolve residue in 10 mL water.

Conditioning:	3 mL methanol (dispensing speed 1 mL/min), 5 mL dist. water (disp. speed 1 mL/min)
Application:	9 mL water sample (disp. speed 3 mL/min over sample loop)
Washing:	10 mL dist. water (disp. speed 3 mL/min)
Drying:	100 mL air (disp. speed 100 mL/min)
Elution:	5 mL ethyl acetate /!methanol (80:20, v/v)
Drying:	100 mL air (disp. speed 100 mL/min)
Evaporation:	under nitrogen, 40 °C
Reconstitution:	in 1 mL dist. water / acetonitrile (95:5, v/v)

The SPE application was performed with a FREESTYLE® SPE automation system.

Further analysis: LC-MS/MS, according to MN Appl. No. 128140

Column:	EC 150/2 NUCLEODUR® π², 5 µm
MN REF:	760624.20
Eluent:	A: dist. water B: acetonitrile 5–95 % B in 7.5 min, 95 % B for 1 min, 95–5 % B in 1 min, 5 % B for 5 min
Flow rate:	0.3 mL/min
Temperature:	35 °C
Detection:	MS, Selected Reaction Monitoring (SRM)
Injection:	5 µL

Recovery rates ± RSD [%], n = 5

Compound	CHROMABOND® HLB	Oasis® HLB
Chloramphenicol-d5	90.9 ± 5.4	90.0 ± 9.3

Good to know

Antibiotics and pesticides contamination of agricultural products such as honey has been an issue in the recent years and resulted in stricter guidelines in food safety control.

*Same conditions for all used columns. Due to a better comparability CHROMABOND® HLB and Oasis® HLB adsorbents (60 µm) were packed into equal column hardware. The shown chromatograms may not be representative of other applications.

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Applications

Pesticides from tap water

MN Appl. No. 306360

Columns*:	CHROMABOND® HLB / 60 µm / 3 mL / 200 mg Oasis® HLB / 60 µm / 3 mL / 200 mg
MN REF:	730924
Conditioning:	5 mL methanol, 5 mL dist. water
Application:	1000 mL tap water (spiked with 50 ng of each analyte)
Washing:	10 mL dist. water
Drying:	5 min with applied vacuum (-15 psi)
Elution:	6 mL acetonitrile
Evaporation:	Under nitrogen, 40 °C
Reconstitution:	In 1 mL dist. water / acetonitrile (95:5, v/v)

Further analysis: LC-MS/MS, according to MN Appl. No. 128150

Column:	EC 50/2 NUCLEOSHELL® PFP, 2.7 µm
MN REF:	763532.20
Eluent:	A: dist. water + 0.1 % formic acid B: acetonitrile + 0.1 % formic acid 5–95 % B in 15 min, 95 % B for 5 min, 95–5 % B in 1 min, 5 % B for 9 min
Flow rate:	0.3 mL/min
Temperature:	40 °C
Detection:	MS, Selected Reaction Monitoring (SRM)
Injection:	5 µL

Recovery rates ± RSD [%], n = 5

Compound	CHROMABOND® HLB	Oasis® HLB
Acetamiprid	73.3 ± 5.0	112.1 ± 9.9
Atrazine	110.3 ± 17.8	114.0 ± 11.6
Azoxystrobin	74.7 ± 5.4	98.1 ± 10.8
Carbaryl	65.7 ± 5.4	69.1 ± 7.1
Chlorotoluron	82.7 ± 5.7	101.2 ± 3.8
Chlorpyrifos	50.3 ± 5.4	47.0 ± 3.7
Clofentezine	27.8 ± 2.7	21.4 ± 3.7
Clothianidin	69.4 ± 6.5	52.9 ± 2.9
Coumaphos	69.8 ± 4.8	82.3 ± 5.2
Cyanazine	99.8 ± 9.3	85.1 ± 7.2
Desethylatrazine	94.8 ± 15.1	87.4 ± 11.4
Desisopropylatrazine	92.5 ± 7.6	N/A
Diazinon	71.5 ± 7.9	73.3 ± 4.7
Difenoconazole	83.9 ± 6.5	28.8 ± 5.0
Diuron	70.0 ± 4.8	80.1 ± 8.4
Ethoprophos	72.4 ± 9.3	85.4 ± 7.2
Hexazinone	88.4 ± 7.7	104.3 ± 7.4
Imazalil	27.3 ± 15.7	N/A
Imidacloprid	93.4 ± 5.1	40.3 ± 5.2
Isoproturon	100.2 ± 4.2	102.8 ± 13.0
Linuron	84.5 ± 7.6	88.3 ± 9.5
Methabenzthiazuron	72.5 ± 5.3	48.0 ± 3.7
Methomyl	78.8 ± 5.4	83.6 ± 5.6
Metobromuron	73.8 ± 5.6	85.6 ± 9.3
Metolachlor	79.0 ± 5.2	89.2 ± 5.0
Monolinuron	75.4 ± 6.2	97.9 ± 7.2
Myclobutanil	101.8 ± 11.4	88.7 ± 14.5
Phosalone	63.8 ± 7.7	74.0 ± 4.0
Piperonylbutoxide	101.4 ± 8.6	99.7 ± 7.9
Propazine	102.1 ± 13.6	90.9 ± 9.4
Propyzamide	84.8 ± 7.1	86.4 ± 10.6
Terbuthylazine	107.9 ± 13.3	100.0 ± 13.6
Thiacloprid	74.1 ± 6.3	86.5 ± 10.8