BioTrap 500 C18/C8

for direct injection of up to 500 µl plasma in HPLC

INSTRUCTION MANUAL

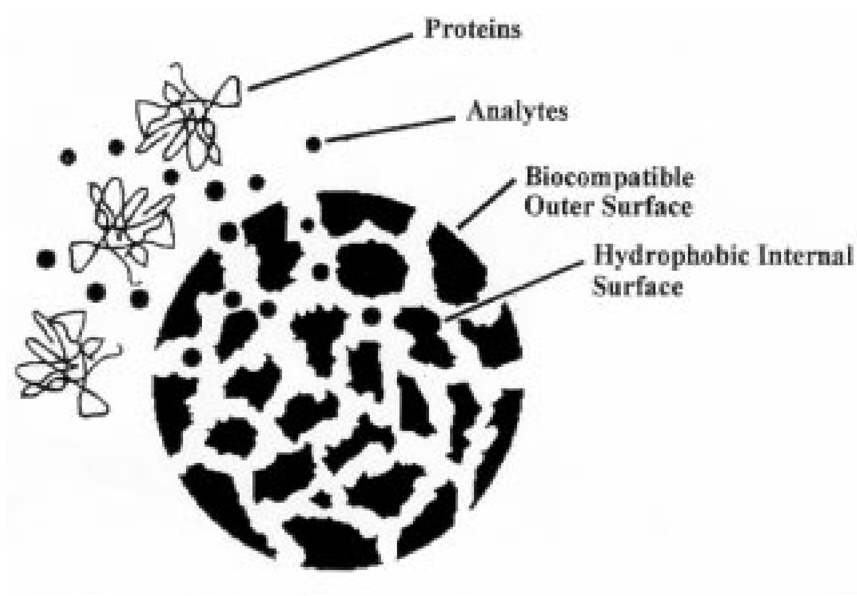

Illustration of a BioTrap 500 particle

ChromTech

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Instruction manual BioTrap 500 C18/C8

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WHAT IS BioTrap 500 C18 and BioTrap 500 C8 ?

BioTrap 500 C18 and BioTrap 500 C8 are new biocompatible sample extraction columns, enabelling repeated direct injections of plasma, serum, milk, supernatants of cell cultures, fermentation broth and other complex matrices, into the HPLC-system without previous cleanup procedures (except for a simple centrifugation).

The BioTrap 500 C18 and 500 C8 are silica based extraction columns with a biocompatible external surface and a hydrophobic internal surface (C18/C8 groups). The biocompatibility has been obtained by attachment of the plasma protein α_1 - acid glycoprotein (AGP) on the external surface of the particles. AGP is an extremely stable protein which tolerates organic solvents used in reversed- phase HPLC. Thee surface within the pores is derivatized with C18/C8 groups and the pores of the matrix are small enough to exclude the plasma proteins and other macromolecular compounds.
Detection methods as UV, fluorescence, electrochemical detection and mass spectrometry can be used. It is favourable to use a detection method giving as high detection selectivity as possible.

SET-UP OF AN HPLC COLUMN SWITCHING SYSTEM

By using a 6-port valve with an electric actuator and injecting the plasma sample with an autosampler, coupled to an extra pump for the extraction mobile phase, it is possible to obtain a complete automatization of the system.

Coupling of the different components to the valve:

A 6-port valve is depicted in Fig. 1. The different components are connected to different ports in the valve as described below:

- Connect the extraction column between **port 3 and 6**
- Connect Pump A to the autosampler and the outlet of the autosampler to the filter. The filter is coupled to **port 1.**
- Connect a tubing from **port 2** to waste.
- Connect Pump B to **port 4.**
- Connect the inlet of the analytical column to **Port 5.**

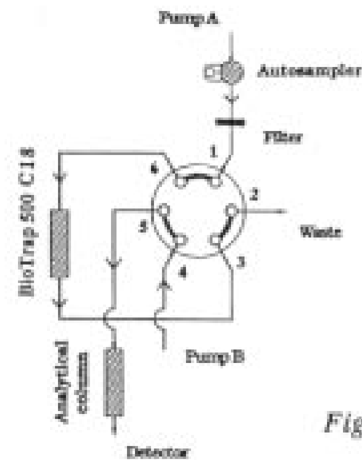


Fig .1

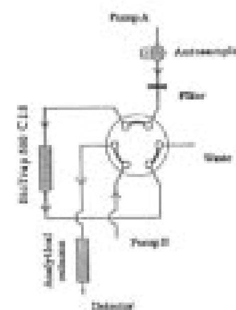
All tubing where plasma is transported, ie.from the autosampler to the extraction column to the waste tubing, should be made of tubing with an ID of at least 0.5 mm.

Description of the column-switching

Pump A is pumping the extraction mobile phase through the autosampler where the sample is injected. After the autosampler a filterholder is inserted, with a 2 µm biocompatible filter. After the passage of the filter, the sample is transported to the extraction column via the 6-port valve **(extraction position). Pump B** is pumping the analytical mobile phase dirough the analytieal column via the 6-port valve **(extraction position).**

In **the elution position** the flow from pump A (the extraction mobile phase) is going to waste. The flow from pump B (the analytical mobile phase) is backflushing the extraction column into the analytical

Extraction position



Elution position

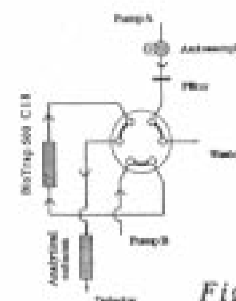


Fig.2

HOW TO USE BIOTRAP 500 C18 / C8

Extraction position

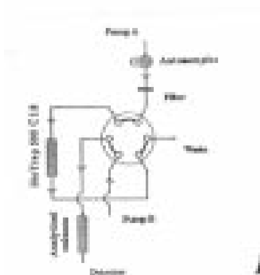


Fig.3

1. **Extraction position - injection and separation of the analyte(s) from high molecular weight compounds (e.g.proteins).**
In this position the sample (e.g. plasma) is injected onto BioTrap 500 C18/C8 (the extraction column). The plasma proteins and other macromolecular compounds will be excluded from the pores and eluted to waste. Low molecular weight compounds penetrate the pores of the particies and are retained by the hydrophobic groups (C18/C8) on the inner surface of the particles. When the proteins have been washed out from the column, the valve can be switched to the elution position.

Elution position

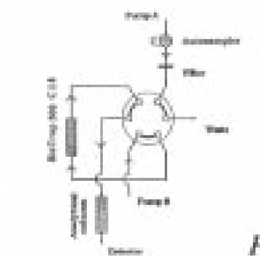


Fig.4

2. Elution position - transfer of the adsorbed compounds from the extraction column to the analytical column. In this position the analytes will be eluted from the extraction column by the analytical mobile phase. When the analytes have been transferred to the analytical column, the valve can be switched back to the extraction position.

Extraction position

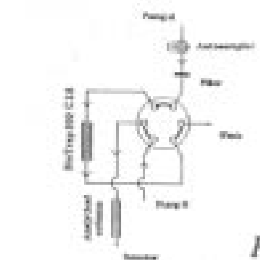


Fig.5

3. Extraction position - the final separation of the analyte(s) on the analytical column and reequilibration of the extraction column. In this position the final separation of the analytes is achieved on the analytical column. Simultaneously, the extraction column is reequilibrated with the extraction mobile phase and the system will be ready for the next injection.

INSTALLATION OF THE BIOTRAP 500 C18 / C8 COLUMN

Wash the column with the analytical mobile phase at a flow rate of 1 ml/min for 10 minutes. Equilibrate the column with the extraction mobile phase for about 5 minutes (1.6 ml/min). When changing the analytical mobile phase it is advised to wash the extraction column for about 30 minutes (1 ml/min) with this mobile phase.
It is recommended to use our special designed 2 µm filter (art.no. F- 117), with filter holder in PEEK (art.no. F-114), coupled on-line in front of the BioTrap 500 C18/C8 column (see Fig. 1), in order to protect the column from particulate impurities.
At least one plasma injection should be made prior to regular use.

<u>Please note that there is a notch in the column under the label. where a wrench can be inserted.</u>

SAMPLE PREPARATION

The samples should be centrifuged before injection to remove cryoproteins and other particulate impurities. After centrifugation the cryoproteins will be present as a white or light yellow layer on top of the plasma. Other particulate impurities may be present in the bottom of the tube.
Note!!! Be sure that the washing liquid in the autosampler does not precipitate the plasma proteins. As for example 5% 2-propanol in distilled water can be used.
The use of a guard column is also recommended in order to protect the analytical column.

EXTRACTION MOBILE PHASE COMPOSITION

The general rule is to choose a pH of the extraction mobile phase which gives the analyte as low charge as possible in order to obtain high recovery. Since the column is silica based the **pH limits is 2.3-7.5.** However, **avoid pH between 3-5** since plasma proteins are more easily precipitated in this range due to the low netcharge of the major plasma proteins in this pH range.
To test the biocompatibility of the extraction mobile phase, mix 0.5 ml of centrifuged plasma/ serum with 0.5 ml of the buffer in a tube. For comparison, make also another sample with 0.5 ml plasma/serum mixed with 0.5 ml distilled water. After 15 minutes the plasma/serum solution should be as clear as before adding the buffer.
Buffer concentrations between 10-100 mM can be used, however 10-50 mM are normally used.

An **organic solvent** should be added to the extraction mobile phase in order to improve the washing of the extraction column and to displace the drug from the plasma protein binding. The solvent can be 2-propanol, acetonitrile ar methanol.
The recovery is affected by the organic solvent concentration. **The recommendation is to start with 4% 2-propanol in the buffer** and then vary to higher and lower concentration for optimization.

Below is the maximum concentrations of three organic solvents that can be used in the extraction mobile phase.

Solvent	Max.	concentradon in the extraction mobile phase
2-propanol	*5%*	(used at pH 2.3-2.6 and pH 6-7.5)
Acetonitrile	10%	(used at pH 6-7.5)
Methanol	10%	(used at pH 6-7.5)

Note that only 2-propanol can be used at low pH!!

If the extraction column is used with a charge organic modifier, this extraction column should only be used with mobile phases containing this charged organic modifier. The reason is that charged organic modifiers may be very difficult to totally remove from the extraction column.

GENERAL METHODS FOR BASIC, ACIDIC AND NON-PROTOLYTIC COMPOUNDS

Below general procedures for basic, acidic and non-protolytic compounds are presented. The methods work for an extremely broad range of compounds.

1. Analysis of basic compounds

Extraction mobile phase:

4% 2-propanol and 5 mM sodium octanesulfonic acid in 30 mM sodium phosphate buffer pH 7.0.

Usually a concentration of 1-10 mM of the ionpairing agent is used.
It is possible to use the column without an ionpairing agent. However, by adding an ionpairing agent (such as sodium octanesulfonic acid) to the extraction mobile phase the recovery of hydrophilic basic analytes can be increased. Furthermore, the separation efficiency is strongly improved.

Analytical mobile phase:

2 mM sodium octanesulfonic acid in an phosphate buffer (pH 2.8 - 3.0) with appropriate amount of organic modifier.

Using an analytical mobile phase with lower concentration of the ionpairing agent, compared to the extraction mobile phase, it is possible to obtain an enrichment effect of the basic analyte on the top of the analytical column, giving an improvement of the separation efficiency.

If MS detection is to be used, a volatile ionpairing reagent like pentafluoropropionic acid can be used in volatile buffers. However, when using MS detection we recommend the use of the BioTrap column designed for MS work, BioTrap 500 MS. See the web site: htip://www.chromtech.se/biotrap or The Application Note no. 16. See also ordering information on the last page.

Comments on the ion-pair technique

In application no. 5 atenolol is extracted and chromatographed using the ionpair technique with BioTrap 500 C18/C8. 200 µl of plasma is injected. If atenolol is extracted on the BioTrap 500 C18/C8 column without the addition of the ionpair agent the recovery is around 50%. When 5 mM sodium octylsulfate is added to the extraction mobile phase, the recovery is increased to 100%.
Application no. 2 shows a chromatogram obtained after online-extraction of 500 µl of plasma, containing 12 ng/ml of propranolol. In this method octanesulfonic acid sodium salt is added to the mobile phases. Compared to a system without ionpairing agent, the separation efficiency is strongly improved.

2. Analysis of acidic compounds

Acidic compounds can be extracted in two ways, either in the uncharged form, or as an ion-pair and by adding a displacement agent like octanoic acid competing with the the analyte for protein binding, giving a larger free fraction that is more easily extracted.

A. Extraction in uncharged form

Extraction mobile phase:

2% 2-propanol in a 50 mM phosphate buffer pH 2.5
The larger plasma volume that is injected, the higher buffer concentration should be used (up to 100 mM). In order to increase the recovery of acidic compounds, the plasma sample could be diluted with a certain volume of the extraction mobile phase prior to centrifugation and injection.

Analytical mobile phase:

Use a method that gives a good chromatographic performance and high sensitivity. Preferably a buffer with pH of about 7.

B. Extraction using the displacement technigue

Extraction mobile phase:

2% 2-propanol and 5 - 10 mM octanoic acid in 30 mM sodium phosphate buffer pH 7.0.

Analytical mobile phase:

Use a method that gives a good chromatographic performance and high sensitivity. Preferably a buffer with pH of about 7.

Comments on the displacement technique

An acid can be extracted in two different ways, as the protonated acid and as an ion-pair. Normally a very good recovery can be obtained by extraction of the protonized acid at low pH. Another way to increase the recovery is to use neutral pH and extract the acid as an ion-pair with positively charged buffer ions like sodium, with a hydrophobic acid (for example octanoic acid) present in the extraction mobile phase. The hydrophobic acid will compete with the analyte for the protein binding and displace the analyte. This can also have a very positive effect on the efficiency. Furthermore, in this way the peaks can be compressed and the recovery can increase. This is very favorable in low concentration analysis. Such results are shown in Application no. 6 for ibuprofen, where 10 µl of serum is injected. The concentration is 20 µg/ml, however, as can be seen from the chromatogram, much lower concentrations can be detected due to the very good chromatographic performance, which means that the method can easily be used for determination of the concentration after single doses of ibuprofen.

3. Analysis of non-protolytic compounds

Extraction mobile phase:

4% 2-propanol in a 30 mM phosphate buffer pH of about 7.

Analytical mobile phase:

Use a method that gives a good chromatographic performance and high sensitivity. Preferably a buffer with pH of about 7.

METHODS USING MASS SPECTROMETRIC DETECTION

If MS detection is to be used, a volatile ionpairing reagent like pentafluoro propionic acid can be used in volatile buffers. However, when using MS detection we recommend the use of the BioTrap column designed for MS work, BioTrap 500 MS. See the web site: http://www.chromtech.se/biotrap or The Application Note no. 16. See also ordering information on the last page.

INJECTION VOLUME, FLOW RATES, EXTRACTION AND ELUTION TIMES

Up to 500 µl of plasma can be repetitively injected. The recovery might be affected by the injection volume. **The 20x4,0 mm is recommended** as the starting column. For smaller sample volumes and if the compound has a low degree of protein binding and/or high affinity to the extraction column the 13x4.0 mm column can be used. When using micro analytical columns the BioTrap micro extraction column (20 x 2.0 mm) can be used.

Extraction

Recommended flow rate is 1.6 ml/min (up to 3.2 ml/min can be used). The extraction time is dependent on the injection volume. Examples of extraction times are given below for plasma/serum injections using an **extraction flow of 1.6 ml/min.**

Injection volume (µl)	*Extraction time (min)*
10 - 25	1 - 2
50 - 100	2 - 3
100 - 250	3 - 4
250 - 500	5

See also applications below.

Elution of the extraction column (backflush)

A flow rate of 1 ml/min is recommended if a 4.6 mm i.d. analytical column is used. The elution time is dependent of injected volume and the eluting strength of the analytical mobile phase. Normally 3-4 minutes elution time is used. See also applications below.

Reequilibration of the extraction column

After elution of a sample and prior to injection of a new sample the extraction column must be equilibrated with the extraction mobile phase. An equilibration time of at least 2-3 minutes is recommended at a flow rate of 1.6 ml/min. See also the “Applications” for different examples.

METHOD DEVELOPMENT

Note! Serum/plasma samples must be centrifuged before injection. See also instructions under “Sample preparation”.

1. Choose detection method.
Decide how to detect the analyte in order to obtain high enough sensitivity and detection selectivity. It is favorable to use methods like fluorescence, electrochemical detection, UV at a selective wavelength or masspectrometric detection, in order to avoid disturbing peaks in the chromatogram. However, for some compounds even UV-detection at 210 nm where very low detection selectivity is obtained, can be used (see phenytoin + carbamazepine and nortriptyline under “Applications”).

2. Develop a preliminary analytical method.
See above under “General methods for basic, acidic and non-protolytic compounds” and develop a preliminary analytical method, by choosing a column and a mobile phase composition giving good chromatographic performance. If the analyte is a basic compound, we recommend the use of ion-pair chromatography. Sodium octane sulfonic acid can be used as counter ion at concentrations between l-l0 mM. This step is performed using only the analytical column. The capacity factor should be in the range of about 2-7 the analyte, according to standard chromatographic theory.

3. Couple the extraction and the analytical column together with the switching valve.
Couple the analytical and the extraction column together with the six-port switching valve according to Figs. 1 and 2. Use the extraction mobile phases recommended above under “General methods for basic, acidic and non-protolytic compounds”.

4. Optimization of the coupled column system
Optimize the analytical method to avoid interferences by changing the mobile phase composition, by changing analytical column to one with different surface chemistry or by changing the detection method. In some cases the extraction mobile phase recommended under “General methods for basic, acidic and non-protolytic compounds” might be slightly changed in order to obtain high enough recovery. This can be done by adjusting the organic modifier concentration or by adjusting the concentration of the ionpairing agent, the buffer concentration or pH.

5. Validate the method.

MAINTENANCE AND COLUMN LIFETIME
The lifetime of the BioTrap 500 Cl8/C8 column is dependent of the sample matrix, the injected volume, the composition of the mobile phases and how the extraction column is rinsed by the extraction mobile phase and by the analytical mobile phase.
Under optimal conditions more than 100 ml of plasma can be injected on the same column.
It is advisable to change the analytical guard column regularly. The filter in front of the extraction column should be exchanged when about 50 ml of serum/plasma have been injected.

STORAGE

The extraction column can be stored at room temperature. When not in use it is recommended to fill the column with 15% 2-propanol in distilled water.

APPENDIX

If the masspectrometer should be used as the detector, we recommend using the BioTrap column designed for MS applications, the BioTrap 500 MS column which can be used in the pH range 2-10. See the website **http://www.chromtech.se/biotrap** where you also can find information and applications of all the BioTrap columns.

Double the speed of bioanalysis using a 10-port switching valve

By using a 10-port valve, faster bioanalysis can be achieved. By connecting two BioTrap 500 columns in a 10-port valve (2-position), it is possible to handle two samples simultaneously. Fig. 6 shows the connection of two BioTrap 500 columns and one analytical column in one HPLC system.
In the first position, the sample is extracted on BioTrap 500 column **1**, while the previous sample is backflushed with the analytical mobile phase from BioTrap 500 column **2** onto the analytical column. In the second valve position, the sample extracted on BioTrap 500 column **1** is backflushed onto the analytical column, while the next sample is injected onto BioTrap 500 column **2** for extraction.

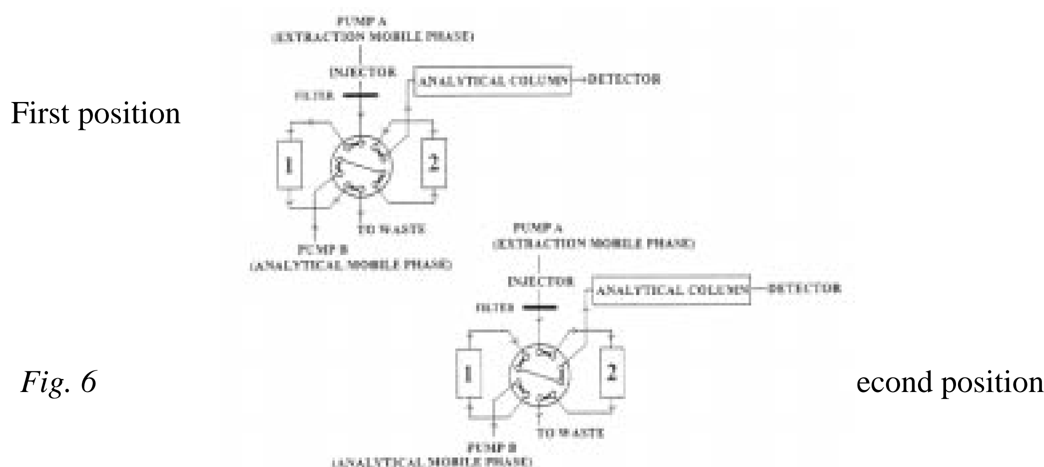
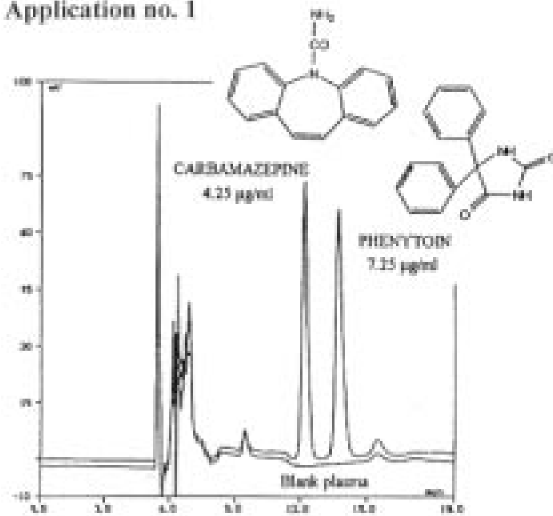


Fig. 6

References

1. J. Hermansson, A. Grahn and I. Hermansson J. Chromatogr., 660/1-2, 119 (1994)
2. J. Hermansson, A. Grahn and I. Hermansson, Current separations , 16. No. 2, 55 (1997)
3. J. Hermansson et al. presented at HPLC 97, Birmingham
4. ChromTech Application Note no. 14
5. ChromTech Application Note no. 15
6. J. Hermansson, A. Grahn and I. Hermansson, J. Chromatog., in press

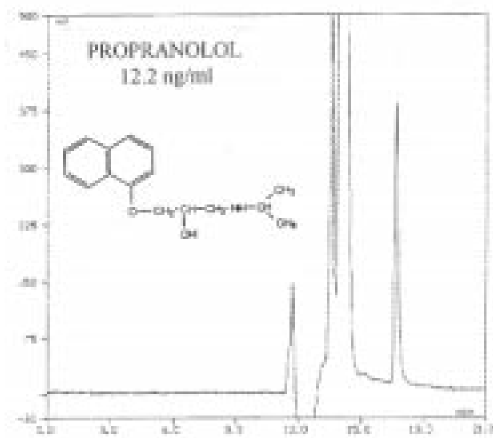
Application no. 1



Inj.vol.: 50 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction):
4% 2-propanol in 20 mM sod. ph.b. pH 6.0
Flow: 0.8 ml/min
Analytical column: Zorbax SB-CN, 150x4.6 mm, 5 µm
Analytical mobile phase:
28% acetonitrile in sod. ph.b. pH 2.8 (µ=0.1)
Flow: 1.0 ml/min
Detection: UV 210 nm
Analysis Program*

Switching valve position	Analysis step	Time
1. Extraction position	sample extraction	4 min
2. Elution position	transfer of analyte	6 min
3. Extraction position	separation and reequilibration	6 min

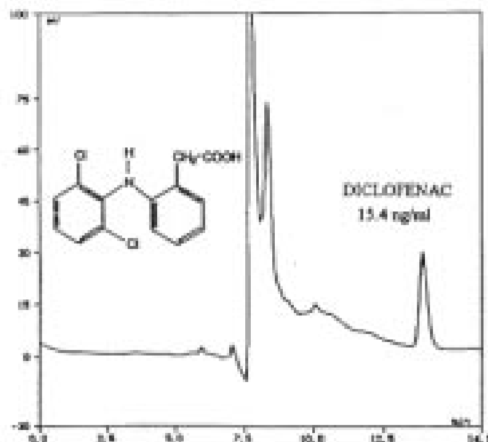
Application no. 2



Inj.vol.: 500 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction):
4% 2-propanol and 5 mM sod.octanesulfonic acid in 20 mM sod. ph.b. pH 7.0
Flow: 0.8 ml/min
Analytical column: Hypersil Elite, 150x4.6 mm, 5 µm + guard Hypersil Elite 10x4.0 mm
Analytical mobile phase:
33% acetonitrile and 2 mM sod.octanesulfonic acid in 116 mM sod. ph.b. pH 2.8
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 220 nm, Em = 340 nm
Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	10 min
2. Elution position	transfer of analyte	6 min
3. Extraction position	separation and reequilibratio	5 min

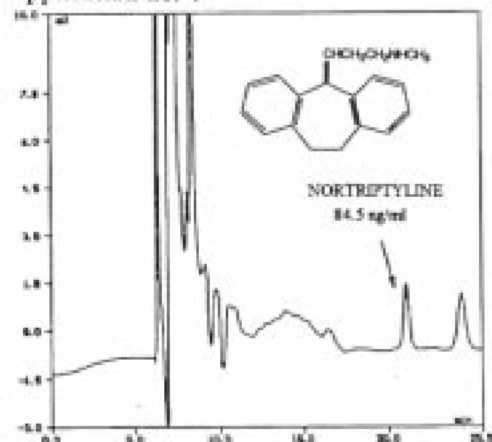
Application no. 3



Inj.vol.: 50 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction):
4% 2-propanol in 50 mM sod. ph.b. pH 5.0
Flow: 0.8 ml/min
Analytical column: Nucleosil C18, 150x4.6 mm, 5 µm + guard CT-sil C18, 10x3.0 mm
Analytical mobile phase:
32% acetonitrile in 42 mM sod. ph.b. pH 6.5
Flow: 0.75 ml/min
Fluorimetric detection: Ex = 290 nm, Em = 360 nm
On-line postcolumn derivatization:
Beam Boost 254 nm, reaction coil 5m x 3mm
Analysis Program*

Switching valve position	Analysis step	Time
1. Extraction position	sample extraction	4 min
2. Elution position	transfer of analyte	6 min
3. Extraction position	separation and reequilibration	6 min

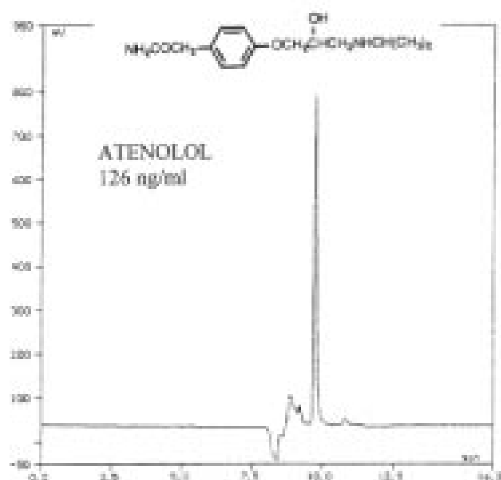
Application no. 4



Inj.vol.: 200 µl serum
Extraction column: BioTrap 500 C18, 13x4.0 mm
Mobile phase (extraction):
4% 2-propanol in 20 mM sod. ph.b. pH 7.0
Flow: 0.8 ml/min
Analytical column: Zorbax Eclipse XDB-C8, 150x4.6 mm, 5 µm
Analytical mobile phase:
28% acetonitrile in sod. ph.b. pH 2.8 (µ=0,1)
Flow: 1.0 ml/min
Detection: UV 210 nm
Analysis Program*

Switching valve position	Analysis step	Time
1. Extraction position	sample extraction	3 min
2. Elution position	transfer of analyte	5 min
3. Extraction position	separation and reequilibration	13 min

Application no. 5

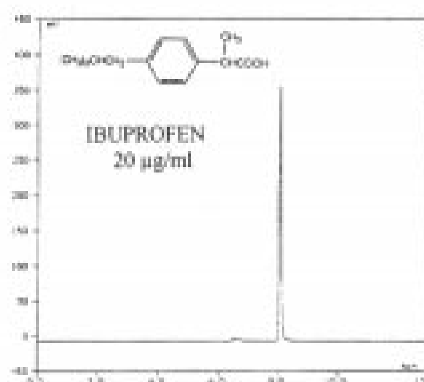


Inj.vol.: 200 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction):
2% 2-propanol and 5 mM sodium octylsulfate in 30 mM sod. ph.b. pH 7.0
Flow: 0.8 ml/min
Analytical column: Zorbax SB-CN, 150x4.6 mm, 5 µm
Analytical mobile phase:
25% acetonitrile and 2 mM sodium octylsulfate in 30 mM sod. ph.b. pH 3.0
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 230 nm, Em = 300 nm

Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	6 min
2. Elution position	transfer of analyte	6 min
3. Extraction position	separation and reequilibratio	4 min

Application no. 6

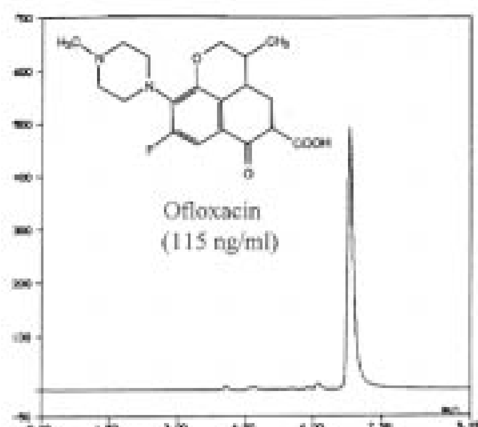


Inj.vol.: 10 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction):
2% 2-propanol and 10 mM octanoic acid in 30 mM sod. ph.b., pH 7.0
Flow: 0.8 ml/min
Analytical column: CT-sil C18, 150x4.6 mm, 5µm
Analytical mobile phase:
35% acetonitrile in 30 mM sod. ph.b. pH 7.0
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 225 nm, Em = 535 nm

Analysis Program*

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	3 min
2. Elution position	transfer of analyte	4 min
3. Extraction position	separation and reequilibratio	4 min

Application no. 7

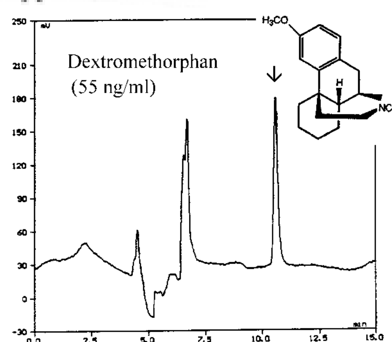


Inj.vol.: 50 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction): 2% 2-propanol and 5 mM sod.octanesulfonic acid in 30 mM sod. ph.b. pH 7.0
Flow: 1.6 ml/min
Analytical column: Zorbax SB-CN, 5 µm, 150x4.6 mm + guard
Analytical mobile phase: 24% acetonitrile and 2 mM sod.octanesulfonic acid in 116 mM sod. ph.b. pH 2.8
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 290 nm, Em = 500 nm

Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	2 min
2. Elution position	transfer of analyte	3.5 min
3. Extraction position	separation and reequilibratio	2 min

Application no. 8

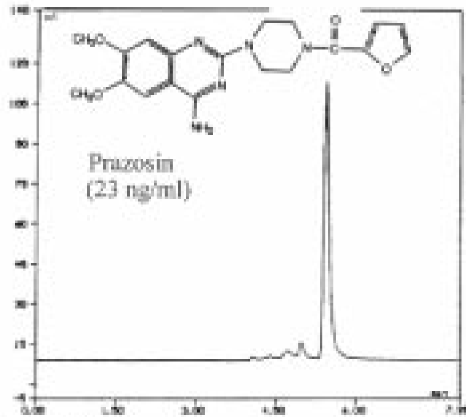


Inj.vol.: 50 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction): 4% 2-propanol and 5 mM sod.octanesulfonic acid in 30 mM sod. ph.b. pH 7.0
Flow: 1.6 ml/min
Analytical column: Zorbax SB-CN, 150x4.6 mm, 5 µm + guard
Analytical mobile phase: 35% acetonitrile and 2 mM sod.octanesulfonic acid in 116 mM sod. ph.b. pH 2.8
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 340 nm, Em = 385 nm

Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	2 min
2. Elution position	transfer of analyte	4 min
3. Extraction position	separation and reequilibratio	2 min

Application no. 9

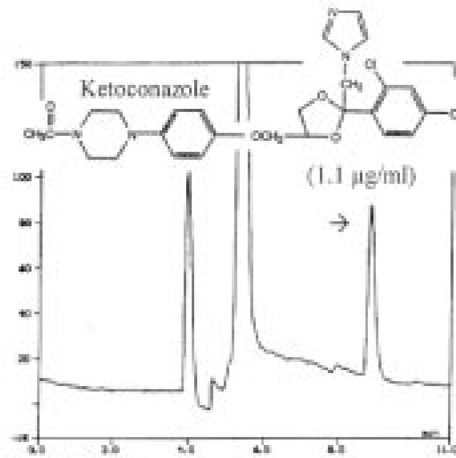


Inj.vol.: 50 µl serum
Extraction column: BioTrap 500 C18, 20x4.0 mm
Mobile phase (extraction): 4% 2-propanol and 5 mM sod.octanesulfonic acid in 30 mM sod. ph.b. pH 7.0
Flow: 1.6 ml/min
Analytical column: Zorbax SB-CN, 150x4.6 mm, 5 µm +guard
Analytical mobile phase: 35% acetonitrile and 2 mM sod.octanesulfonic acid in 116 mM sod. ph.b. pH 2.8
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 340 nm, Em = 385 nm

Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	2 min
2. Elution position	transfer of analyte	4 min
3. Extraction position	separation and reequilibratio	2 min

Application no. 10



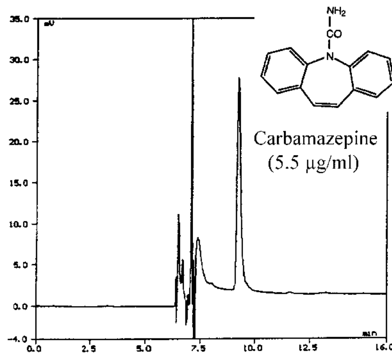
Inj.vol.: 200 µl serum
Extraction column: BioTrap 500 C8, 20x4.0 mm
Mobile phase (extraction): 4% 2-propanol and 5 mM sod.octanesulfonic acid in 20 mM sod. ph.b. pH 7.0
Flow: 1.6 ml/min
Analytical column: Kromasil C18, 5 µm, 100x4.6 + guard
Analytical mobile phase: 35% acetonitrile and 2 mM sod.octanesulfonic acid in 116 mM sod. ph.b. pH 2.8
Flow: 1.0 ml/min
Fluorimetric detection: Ex = 220 nm, Em = 370 nm

Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	3 min
2. Elution position	transfer of analyte	5 min
3. Extraction position	separation and reequilibratio	3 min

Note that a Micro BioTrap 500 column has been used for the application below.

Application no. 11



Inj.vol.: 10 µl serum
Extraction column: Micro BioTrap 500 C18, 20x2.0 mm
Mobile phase (extraction): 4% 2-propanol in 20 mM sod. ph.b. pH 6.0
Flow: 0.2 ml/min
Analytical column: Micro CT-sil C18, 150x2.0 mm, 5 µm + guard
Analytical mobile phase: 65% methanol in 116 mM sod. ph.b. pH 2.8
Flow: 0.2 ml/min
Detection: UV 285 nm

Analysis Program

Switching valve positio	Analysis step	Time
1. Extraction position	sample extraction	5 min
2. Elution position	transfer of analyte	5 min
3. Extraction position	separation and reequilibratio	6 min

Ordering information-BioTrap 500 C18/C8

Part no.	Description	Part no.	Description
B18134K	BioTrap 500 C18, 13x4.0mm, complete with holder	B8134K	BioTrap 500 C8, 13x4.0mm. , complete with holder
B18204K	BioTrap 500 C18, 20x4.0mm, complete with holder	B8204K	BioTrap 500 C8, 20x4.0mm, complete with holder
B18134C	BioTrap 500 C18, 13x4.0mm, 2 cartridges	B8134C	BioTrap 500 C8, 13x4.0mm, 2 cartridges
B18204C	BioTrap 500 C18. 20x4.0mm, 2 cartridges	B8204C	BioTrap 500 C8, 20x4.0mm, 2 cartridges
B18202K	BioTrap 500 C18, 20x2.0mm, complete with holder	B8202K	BioTrap 500 C8, 20x2.0mm, complete with holder
B18202C	BioTrap 500 C18, 20x2.0mm, 2 columns	B8202C	BioTrap 500 C8, 20x2.0mm, 2 columns

Part no.	Description
KIT - 1	BioTrap 500 C18. 13x4.0mm + 20x4.0mm + holder
KIT - 2	BioTrap 500 C18. 13x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 3	BioTrap 500 Cl8, 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 4	BioTrap 500 Cl8, 13x4.0mm + 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 5	BioTrap 500 C8. 13x4.0mm + 20x4.0mm + holder
KIT - 6	BioTrap 500 C8. 13x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 7	BioTrap 500 C8. 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 8	BioTrap 500 C8. 13x4.0mm + 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 9	BioTrap 500 C18, 20x4.0mm + BioTrap 500 C8. 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 10	BioTrap 500 MS, 13x4.0mm + 20x4.0mm + holder
KIT - 11	BioTrap 500 MS. 13x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 12	BioTrap 500 MS, 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 13	BioTrap 500 MS. 13x4.0mm + 20x4.0mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 14	BioTrap 500 MS. 20x4.0 mm + BioTrap 500 C18, 20x4.0 mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 15	BioTrap 500 MS. 20x4.0 mm + BioTrap 500 C8. 20x4.0 mm + holder + filterholder in PEEK + 5 PEEK-filters
KIT - 16	BioTrap 500 MS. 20x4.0 mm + BioTrap 500 C18. 20x4.0mm + BioTrap 500 C8. 20x4.0 mm + holder + filterholder in PEEK + 5 PEEK-filters
CH2	BioTrap holder with end fittings, 2 mm ID
CH4	BioTrap holder with end fittings. 4 mm ID
F - 117	PEEK-filter (between autosampler and valve) 2µm, 5pcs
F - 114	Filter holder in PEEK
E - 6pv	6-port valve, conical rotor. fast elektric actuator. 1/16"
E - 6cv	6-port valve, flat rotor. fast elektric actuator. 1/16"

Ordering information-BioTrap 500 MS

Part no.	Description
BMS134K	BioTrap 500 MS, 13x4.0mm, complete with holder
BMS134C	BioTrap 500 MS, 13x4.0mm, 2 cartridges
BMS204K	BioTrap 500 MS, 20x4.0mm, complete with holder
BMS204C	BioTrap 500 MS, 20x4.0mm, 2 cartridges
BMS202K	BioTrap 500 MS, 20x2.0mm, complete with holder
BMS202C	BioTrap 500 MS 20x2.0mm, 2 cartridges