

This Data Sheet Contains Important Information About This Product.

SupelMIP™ SPE – Beta-blockers

Product Description:

Molecular imprinted polymers (MIPs) are a class of highly cross-linked polymer-based molecular recognition elements engineered to bind one target compound or a class of structurally related target compounds with high selectivity. Selectivity is introduced during MIP synthesis in which a template molecule, designed to mimic the analyte, guides the formation of specific cavities or imprints that are sterically and chemically complementary to the target analyte(s). It is therefore critical for analysts to use the methodology described below when using this phase. Conventional generic methodologies employed with conventional SPE chemistries (e.g., reversed-phase C18) will yield sub-optimal results when employed with this phase.

The following methods have been developed for the selective extraction of beta-blockers from both biological matrices and natural water. The methods are highly reproducible and offer beta-blocker recoveries of $\geq 80\%$. The method minimizes matrix effects and offers limits of detection of less than 5 ppt in water and less than 10 ppt for plasma and urine. Since the methods are amenable to the extraction of a wide range of beta-blockers, recoveries may vary for each specific molecule. It is recommended to use the prescribed method as a screening tool to identify which beta-blockers are present. Once specific beta-blockers are identified, conditioning, wash, and elution steps can be further optimized to offer higher recoveries if required.

Extraction Procedure: A flow rate of ~0.5 mL/min. is recommended. For analyte elution a flow rate of ~0.2 mL/min. is recommended.

Application Name:	Trace level extraction of Beta-blockers from natural water ¹	Extraction of Beta-blockers from urine and other biological fluids
Analyte:	Beta-blockers	Beta-blockers
Sample Matrix:	Natural water	Urine or plasma
General Comments:	Typical recoveries are over 80% for atenolol, betaxolol, carazolol, metoprolol, pindolol, propranolol, sotalol and timolol.	Typical recoveries are over 85% for metoprolol, propranolol, carzalolol and atenolol.
SupelMIP SPE – Beta-blockers:	25 mg/10 mL (LRC) (Cat. No. 53218-U); or 25 mg/3 mL (Cat. No. 53213-U)	25 mg/10 mL (LRC) (Cat. No. 53218-U); or 25 mg/3 mL (Cat. No. 53213-U)
Sample Pre-treatment:	None	Urine or plasma (centrifuged at 3000 x g for 10 min.) diluted 1:1 (v/v) with 25 mM ammonium acetate (NH ₄ Ac), pH 5
1. Condition/equilibrate cartridge with:	<ul style="list-style-type: none"> ◆ 1 mL methanol ◆ 1 mL DI water 	<ul style="list-style-type: none"> ◆ 1 mL methanol ◆ 1 mL DI water ◆ 1 mL 25 mM ammonium acetate (NH₄Ac), pH 5
2. Load sample: Note: recommended flow rate is 3 mL/min for natural water, and ~0.5 mL/min. for urine/plasma	Apply 100 mL water sample to the cartridge, pH 5-7. To increase head space volume, stack an empty 60 mL cartridge (Cat. No. 57022) on top of a 3 mL SupelMIP SPE cartridge using an SPE tube adapter (57020-U). For Waste water extractions apply 25 mL of sample.	Apply up to 10 mL diluted urine or 5 mL plasma sample to the cartridge
3. Wash (interference elution): Note: Apply gentle vacuum between each wash step.	<ul style="list-style-type: none"> ◆ 2 x 1 mL DI water (selective elution/removal of salts and hydrophilic matrix components) ◆ Apply full vacuum through cartridge for 2 min. to remove residual moisture from cartridge. ◆ 1 mL acetonitrile (selective removal of hydrophobic interferences) ◆ Apply full vacuum through cartridge for 10 min. to remove residual solvent from cartridge. ◆ 1 mL dichloromethane (to selectively enhance MIP interaction with beta-blockers) ◆ Apply full vacuum through cartridge for 2 min. to remove residual solvent from cartridge. 	<ul style="list-style-type: none"> ◆ 1 mL 50 mM ammonium acetate (NH₄Ac), pH 6.5 ◆ 1 mL DI water (selective elution/removal of salts and hydrophilic matrix components) ◆ Apply full vacuum through cartridge for 2 min. to remove residual moisture from cartridge. ◆ 1 mL acetonitrile (selective removal of hydrophobic interferences) ◆ 1 mL 60% acetonitrile/40% DI Water (selective removal of hydrogen bonded interferences) ◆ Apply full vacuum through cartridge for 10 min. to remove residual solvent from cartridge. ◆ 1 mL dichloromethane (to selectively enhance MIP interaction with beta-blockers) ◆ Apply full vacuum through cartridge for 2 min. to remove residual solvent from cartridge.

4. Analyte elution:
Note: recommended flow rate ~0.2 mL/min.

For 10 mL cartridges, elute beta-blockers with 2 x 1 mL 10% acetic acid in methanol. For 3 mL cartridges, elute beta-blockers with 3 x 1 mL 10% acetic acid in methanol. Apply a gentle vacuum between each fraction. Evaporate and reconstitute with LC mobile phase prior to analysis.

Recommended Analytical Technique: LC-MS

Standard Conditions:

column: Ascentis C18, 5 cm x 2.1 mm I.D., 5 µm particle size (53822-U)
instrument: Waters Micromass ZQ
mobile phase A: 10 mM ammonium acetate (pH unadjusted) in 10% acetonitrile
mobile phase B: acetonitrile
flow rate: 1 mL/min., split to MS
temp.: 35 °C
det.: MS, ESI(+) in selected ion recording (SIR)
injection: 10 µL
gradient:

Min	%A	%B
0.00	100	0
1.00	100	0
15.00	0	100
16.00	100	0

Peak ID:
1. atenolol (M+H)+ : 267.16
2. pindolol (M+H)+ : 249.15
3. timolol (M+H)+ : 317.15
4. metoprolol (M+H)+ : 268.18
5. propranolol (M+H)+ : 260.15
6. betaxolol (M+H)+ : 308.21

Trace Conditions:

column: Ascentis Express C18, 5 cm x 2.1 mm I.D., 2.7 µm particle size (53822-U)
instrument: Applied Biosystems 3200 Q-TRAP MRM
mobile phase: 10 mM ammonium acetate (pH unadjusted) in 10% acetonitrile:acetonitrile (74:26)
flow rate: 0.2 mL/min.
temp.: 35 °C
det.: MS/MS
ion mode: positive
ion source: turbospray
ionspray voltage: 3400 V
source temp.: 375 °C
collision gas: 45 psi
injection: 5 µL

MRM transitions:
1. atenolol (267.27/145.20)
2. pindolol (249.15/116.20)
3. timolol (317.23/261.20)
4. metoprolol (268.29/126.10)
5. propranolol (260.12/116.20)
6. betaxolol (309.00/116.20)

Product Information:**Description****SupelMIP SPE - Clenbuterol**

25 mg/10 mL (LRC)

Pkg. Qty. Cat. No.50 **53201-U****SupelMIP SPE - Beta-agonists (class selective)**

25 mg/10 mL (LRC)

50 **53202-U**

25 mg/3 mL

50 **53225-U****SupelMIP SPE – NNAL**

25 mg/10 mL (LRC)

50 **53206-U**

25 mg/3 mL

50 **53203-U****SupelMIP SPE - Riboflavin (Vitamin B2)**

25 mg/10 mL (LRC)

50 **53207-U****SupelMIP SPE - Triazine 10**

25 mg/10 mL (LRC)

50 **53208-U****SupelMIP SPE - Chloramphenicol**

25 mg/10 mL (LRC)

50 **53210-U**

25 mg/3 mL

50 **53209-U****SupelMIP SPE - Beta-blocker (class selective)**

25 mg/10 mL (LRC)

50 **53218-U**

25 mg/3 mL

50 **53213-U****SupelMIP SPE - TSNAs (NNK, NNN, NAB, NAT)**

50 mg/10 mL (LRC)

50 **53221-U**

50 mg/3 mL

50 **53222-U****SupelMIP SPE - Full Beta-receptors (beta-blockers & beta-agonists)**

25 mg/10 mL (LRC)

50 **53223-U**

25 mg/3 mL

50 **53224-U**

1. Procedure based on worked conducted by Prof. Damia Barcelo et al at the Department of Environmental Chemistry, IIQAB-CSIC, Barcelona, Spain.

SupelMIP SPE developed by MIP Technologies AB

SupelMIP is a trademark of Sigma-Aldrich Co.