



AFFINIMIP[®] SPE

Glyphosate



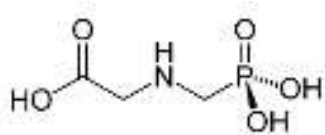
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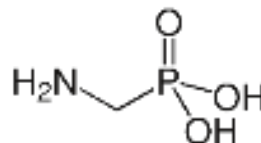
This booklet describes different applications of **AFFINIMIP® SPE Glyphosate** for the analysis of **Glyphosate, AMPA and Glufosinate**. The determination of very low concentrations of these molecules in various waters as well as in very complex matrices is shown.

In addition, as these molecules can be found in surface and underground water, an application with **AFFINIMIP® POCIS Glyphosate** shows the uptake of these molecules as a POCIS passive sampler.

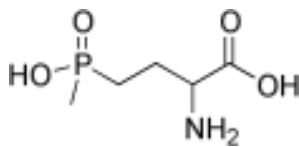
For the analysis of these compounds, several analytical methods have been used such as LC-MS/MS even without derivatization as well as Capillary Electrophoresis –UV.



Glyphosate



AMPA



Glufosinate

Do you know?

Glyphosate and Glufosinate are closely related herbicides referred to as phospho-herbicides. Glyphosate undergoes rapid microbial degradation in plants, soil and water to the metabolite aminomethylphosphonic acid (AMPA). Codex alimentarius has defined a MRL (maximum residue limit) for Glyphosate of 0.05mg/Kg in meat or milk and 30mg/Kg in cereals and for Glufosinate, 2mg/kg of soybean.

- ✓ Efficient for Glyphosate, AMPA, Glufosinate et others metabolites
- ✓ Ready to use kit
- ✓ Tested with LC-MS/MS, Capillary Electrophoresis - UV
- ✓ Do not require derivatization of these molecules
- ✓ Tested on large volume of water, tea, cereals, honey...
- ✓ Simple & Fast process
- ✓ High capacity cartridges



GLYPHOSATE, AMPA, GLUFOSINATE IN CEREALS NO DERIVATIZATION – LC-MS/MS



PROTOCOL OF PURIFICATION

Sample preparation

Mix 3g of crushed cereals + 25mL water with 1% formic acid. Sonicate 30 min, centrifuge 10 min. The supernatant is put to pH = 7 with ammonia solution and filtered to form the loading solution.



Purification with a 6mL AFFINIMIP® SPE Glyphosate cartridge

Equilibration

9mL pure Water

Loading

9mL loading solution

Washing of interferences

24mL Water

Elution (E)

8 mL water with HCl 0.1M

Elutions are evaporated under vacuum at 60°C for 2 hours and dissolved in 3 mL of mobile phase containing EDTA-Na2 0,8mM.

RESULTS

Recovery of Glyphosate, AMPA and Glufosinate after AFFINIMIP® SPE Glyphosate clean-up of Cereals spiked at 92µg/Kg each. No derivatization was performed.

	Glyphosate	AMPA	Glufosinate
Yield (%)	101 %	98 %	93 %
RSD (n=3)	3 %	2 %	3 %

HPLC Method with LC – MS/MS

Analysis by HPLC – MS/MS (QTRAP 4000)

Column: Acclaim trinity Q1 100mm x 3mm ID (3µm)

Mobile phase: gradient with Ammonium formiate 50 mM pH = 2.9 (A) – Acetonitrile (B)

Time (min)	% A	% B
0	100	0
3	100	0
3,2	0	100
6,0	0	100
6,2	100	0
10,2	100	0

Flow rate: 0,5mL/min

Injection volume: 20µL.

Catalog number:

6mL format with enhanced performances

FS113-15-02B for 25 cartridges

FS113-15-03B for 50 cartridges

12mL format

FS113-02C for 25 cartridges

FS113-03C for 50 cartridges

PROTOCOL OF PURIFICATION

Sample preparation

A mixture of 5mL of apple juice + 15mL water + 200µL Formic acid is put to pH = 7 with ammonia solution to form the loading solution.

Purification with a 6mL AFFINIMIP® SPE Glyphosate cartridge

Equilibration

6mL pure Water

Loading

6mL loading solution

Washing of interferences

12mL Water

Elution (E)

8 mL water with HCl 0.1M

Elutions are evaporated under vacuum at 60°C for 2 hours and dissolved in 1 mL of mobile phase containing EDTA-Na2 0,8mM.



RESULTS

Recovery of Glyphosate, AMPA and Glufosinate after AFFINIMIP® SPE Glyphosate clean-up of Apple juice spiked at 67µg/Kg each. No derivatization was performed.

	Glyphosate	AMPA	Glufosinate
Yield (%)	96 %	86 %	92 %

HPLC Method with LC – MS/MS

Analysis by HPLC – MS/MS (QTRAP 4000)

Column: Acclaim trinity Q1 100mm x 3mm ID (3µm)

Mobile phase: gradient with Ammonium formiate 50 mM pH = 2.9 (A) – Acetonitrile (B)

Time (min)	% A	% B
0	100	0
3	100	0
3,2	0	100
6,0	0	100
6,2	100	0
10,2	100	0

Flow rate: 0,5mL/min

Injection volume: 20µL.

Catalog number:

6mL format

FS113-02B for 25 cartridges

FS113-03B for 50 cartridges

12mL format

FS113-02C for 25 cartridges

FS113-03C for 50 cartridges

GLYPHOSATE, AMPA, GLUFOSINATE in BLACK TEA NO DERIVATIZATION – LC-MS/MS



PROTOCOL OF PURIFICATION

Sample preparation

Mix 3g of dry tea +50mL water with 1% formic acid. Sonicate 30 min, centrifuge 10 min. The supernatant is filtered and put to pH = 7 with ammonia solution to form the loading solution.

Purification with a 6mL AFFINIMIP® SPE Glyphosate cartridge

Equilibration

6mL pure Water

Loading

3mL loading solution

Washing of interferences

12mL Water

Elution (E)

8mL water with HCl 0.1M

Elutions are evaporated under vacuum at 60°C for 2 hours and dissolved in 3 mL of mobile phase containing EDTA-Na2 0,8mM.

HPLC Method with LC – MS/MS

Analysis by HPLC – MS/MS (QTRAP 4000)

Column: Acclaim trinity Q1 100mm x 3mm ID (3µm)

Mobile phase: gradient with Ammonium formiate 50 mM pH = 2.9 (A) – Acetonitrile (B)

Time (min)	% A	% B
0	100	0
3	100	0
3,2	0	100
6,0	0	100
6,2	100	0
10,2	100	0

Flow rate: 0,5mL/min

Injection volume: 20µL.



RESULTS

Recovery of Glyphosate, AMPA and Glufosinate after AFFINIMIP® SPE Glyphosate clean-up of flavoured Black Tea spiked at 1,67mg/kg each. No derivatization was performed.

	Glyphosate	AMPA	Glufosinate
Yield (%)	103 %	81 %	100 %
RSD (n=3)	10 %	12 %	6 %
Matrix effect	-3 %	23 %	6%

Catalog number:

6mL format

FS113-02B for 25 cartridges

FS113-03B for 50 cartridges

12mL format

FS113-02C for 25 cartridges

FS113-03C for 50 cartridges

PROTOCOL OF PURIFICATION

Sample preparation

10 grams of honey are mixed with 30 mL of ultrapure water - 1% formic acid and magnetically stirred during 30 minutes. The mixture is then filtered through 0.45µm filter and neutralized to pH 7 with ammonia solution to form the loading solution.



Purification with a 6mL AFFINIMIP® SPE Glyphosate cartridge

Equilibration

6mL pure Water

Loading

3mL loading solution

Washing of interferences

12mL Water

Elution (E)

8mL water with HCl 0.1M

Elutions are evaporated under vacuum at 60°C for 2 hours and dissolved in 3 mL of mobile phase containing EDTA-Na2 0,8mM.

HPLC Method with LC – MS/MS

Analysis by HPLC – MS/MS (QTRAP 4000)

Column: Acclaim trinity Q1 100mm x 3mm ID (3µm)

Mobile phase: gradient with Ammonium formiate 50 mM pH = 2.9 (A) – Acetonitrile (B)

Time (min)	% A	% B
0	100	0
3	100	0
3,2	0	100
6,0	0	100
6,2	100	0
10,2	100	0

Flow rate: 0,5mL/min

Injection volume: 20µL.

RESULTS

Recovery of Glyphosate, AMPA and Glufosinate after AFFINIMIP® SPE Glyphosate clean-up of Honey spiked at 400 µg/kg each. No derivatization was performed.

	Glyphosate	AMPA	Glufosinate
Yield (%)	104 %	89 %	100 %
RSD (n=3)	4 %	8 %	8 %
Matrix effect	-16 %	-36 %	-19 %

Catalog number:

6mL format

FS113-02B for 25 cartridges

FS113-03B for 50 cartridges

12mL format

FS113-02C for 25 cartridges

FS113-03C for 50 cartridges

PROTOCOL OF PURIFICATION

Sample preparation

3g Dried and crushed *Datisca Cannabina* cannabis like plant mixed with 60 mL water - 1% Formic acid were sonicated for 30 mn and centrifuged at 4000 RPM for 10 mn. After filtration, the solution is neutralized at pH = 7 with ammonia solution. The loading solution is obtained after a 0.45µm filtration.



Purification with a 6mL

AFFINIMIP® SPE Glyphosate cartridge

Equilibration

9mL pure Water

Loading

6mL loading solution

Washing of interferences

24mL Water

Elution (E)

8 mL water with HCl 0.1M

Elutions are evaporated under vacuum at 60°C for 2 hours and dissolved in 3 mL of mobile phase containing EDTA-Na2 0,8mM.

RESULTS

Recovery of Glyphosate, AMPA and Glufosinate after AFFINIMIP® SPE Glyphosate clean-up of dried plant spiked at 333µg/Kg each. No derivatization was performed.

	Glyphosate	AMPA	Glufosinate
Yield (%)	70 %	72 %	81%
RSD (n=3)	5 %	7 %	6 %

Catalog number:

AFFINIMIP® SPE Glyphosate

6mL format with enhanced performances

FS113-15-02B for 25 cartridges

FS113-15-03B for 50 cartridges

12mL format

FS113-02C for 25 cartridges

FS113-03C for 50 cartridges

Analytical conditions according to p 16

GLYPHOSATE, AMPA, GLUFOSINATE IN WATER NO DERIVATIZATION – LC-MS/MS



PROTOCOL OF PURIFICATION

Purification with a 6mL AFFINIMIP® SPE Glyphosate cartridge

Equilibration

6mL pure Water

Loading

100mL spiked water

Washing of interferences

6mL Water

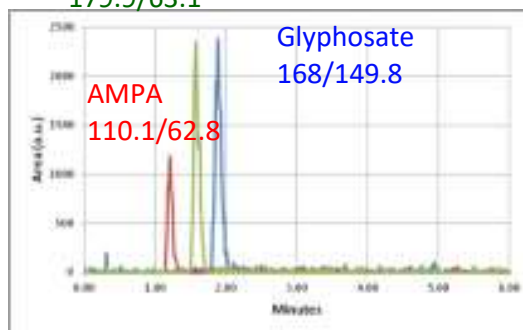
Elution (E)

8mL water with HCl 0.1M

Elutions are evaporated under vacuum at 60°C for 2 hours and dissolved in 3 mL of mobile phase containing EDTA-Na₂ 0,8mM.

RESULTS

Glufosinate
179.9/63.1



Chromatograms obtained after a clean-up with **AFFINIMIP®SPE Glyphosate** for 100mL water spiked at 3µg/L of each molecule

Recovery of Glyphosate, AMPA and Glufosinate after **AFFINIMIP® SPE Glyphosate** clean-up of water spiked at 3µg/L each. No derivatization was performed.

HPLC Method with LC – MS/MS

Analysis by HPLC – MS/MS (QTRAP 4000)

Column: Acclaim trinity Q1 100mm x 3mm ID (3µm)

Mobile phase: gradient with Ammonium formiate 50 mM pH = 2.9 (A) – Acetonitrile (B)

Time (min)	% A	% B
0	100	0
3	100	0
3,2	0	100
6,0	0	100
6,2	100	0
10,2	100	0

Flow rate: 0,5mL/min
Injection volume: 20µL.

	Glyphosate	AMPA	Glufosinate
Yield (%)	91 %	70 %	80 %
RSD (n=3)	2 %	1 %	6 %

Catalog number:

6mL format with enhanced performances

FS113-15-02B for 25 cartridges

FS113-15-03B for 50 cartridges

12mL format

FS113-02C for 25 cartridges

FS113-03C for 50 cartridges

Efficient clean-up and enrichment

PROTOCOL OF PURIFICATION

Sample preparation

Purification with a 6mL AFFINIMIP® SPE Glyphosate cartridge

Equilibration

6mL pure Water

Loading

3 to 500 mL

Washing of interferences

3mL Water

Elution (E)

4-8 mL water with HCl 0.1M

CE analysis (no derivatization)

Column: fused-silica capillary of 60.2 cm (effective length, 50 cm) x 50 µm ID at 25°C

Mobile phase: 7.5 mM phthalic acid - 51.3 mM histidine running buffer (pH 6.5, ionic strength of 21.8 mM, buffer capacity 25 mM L⁻¹ pH⁻¹) containing 1 mM CTAB

Voltage : +25kV

Detection UV-DAD (240nm)

RESULTS

Recovery yields of glyphosate and AMPA after AFFINIMIP® SPE **Glyphosate** clean-up of mineral water spiked at 25µg/mL.

Loading volume 3mL

Analysis done by CE without derivatization

Analytes	Recoveries %
Glyphosate	85
AMPA	87

Publication:

Preliminary recovery study of a commercial molecularly imprinted polymer for the extraction of glyphosate and AMPA in different environmental waters using MS, B. Claude, C. Berho, S. Bayouh, L. Amalric, E. Coisy, R. Nehmé, P. Morin, *Environ Sci Pollut Res*, 24: 12293 (2017).

Catalog number:

3mL format

FS113-02.IP for 25 cartridges

FS113-03.IP for 50 cartridges

6mL format

FS113-02B for 25 cartridges

FS113-03B for 50 cartridges

**Performance not affected by physico chemical properties of Water
Loading with up to 1L**

Physico chemical properties of tested waters

Salt concentrations (mg/L) and pH of analyzed solutions

	Ca	Na	Mg	K	HCO ₃	Cl	NO ₃	SO ₄	Fe	pH
Groundwater	15,7	11,3	4,9	1,3	76	9,7	<0,5	1,2	7,5	7,1
Groundwater	22,3	105,7	17	4,7	136	159	8,9	15,8	0,17	6,4
Groundwater	104,1	13,9	6,9	1,8	203	28,1	113,7	33		7,1
Geothermal water	799	5163,5	189,5	71,9		9759,7		702,2	3,2	
Mineral water	80	6,5	24	1	360	3,8	3,7	12,6		7,2

AFFINIMIP® SPE Glyphosate performance for tested waters

Above five waters spiked at various concentrations with AMPA and Glyphosate

Sample	Concentration range	Average Recoveries %
Glyphosate	100 to 750ng/L	>70%
AMPA	100 to 750ng/L	>75%

Method UPLC – MS/MS

Column: UPLC HSS T3 (2.1mm x 100mm, 1,8µm)

Mobile phase: A: Water/Ammonium

Acetate 5mM

B: Acetonitrile

Time (min)	A %	B %
0	90	10
2	90	10
7	50	50
7.5	0	100
11	0	100

Flow rate: 0.2mL/min

MS detection: m/z 321 (ESI⁻)

Injection volume: 20µL.

Same protocol than previous page

Publication:

B. Claude, C. Berho, S. Bayoudh, L. Amalric, E. Coisy, R. Nehmé, P. Morin, *Environ Sci Pollut Res*, 24: 12293 (2017).

Catalog number:

3mL format

FS113-02.IP for 25 cartridges

FS113-03.IP for 50 cartridges

6mL format

FS113-02B for 25 cartridges

FS113-03B for 50 cartridges

RESULTS

Laboratory sampling rates estimation for AMPA and glyphosate using the AFFINIMIP® POCIS Glyphosate



AFFINIMIP® POCIS Glyphosate

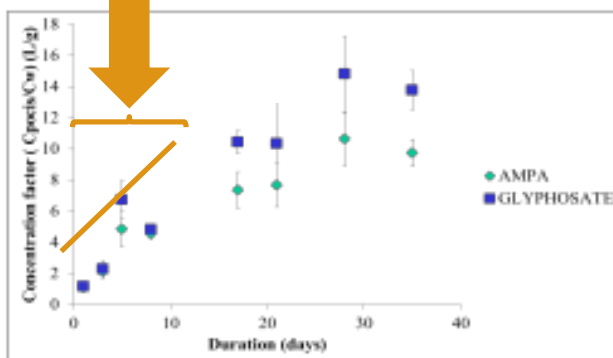
Passive Sampling with POCIS

Polar Organic Chemical Integrative Sampler (POCIS) is a passive sampler designed to provide the time weighted average (TWA) concentration of chemicals during a sampling period of several weeks.

AFFINIMIP® POCIS Glyphosate enables the sampling of Glyphosate and AMPA in water (Groundwater, geothermal, mineral...).

Then the powder is collected in an empty SPE column for the extraction of Glyphosate and AMPA

Sampling rates: 130mL/day/200mg AFFINIMIP® POCIS Glyphosate in agreement with other pesticides in classical POCIS.



Mineral water (pH = 7) fortified at 500ng/L of AMPA and glyphosate. Concentrations kept constant during whole experiment. Pesticides concentration in the tank, temperature, TOC and conductivity monitored during the experimental period to verify the stability of physico-chemical conditions in water.

PROTOCOL OF EXTRACTION

Extraction of collected Glyphosate and AMPA from AFFINIMIP® POCIS Glyphosate with a SPE

Extraction of the analytes (E)

HCl solution (100mM)

The extraction solution is then evaporated and reconstituted with water prior analysis

Publications:

Laboratory calibration of a POCIS-like sampler based on molecularly imprinted polymers for glyphosate and AMPA sampling in water, C. Berho, B. Claude, E. Coisy, A. Togola, S. Bayouth, P. Morin, L. Amalric, *Anal Bioanal Chem* 409: 2029 (2017)

Catalog number:
POCIS-GLY.90.55.A.1 for 1 AFFINIMIP® POCIS Glyphosate
POCIS-GLY.90.55.A.10 for 10 AFFINIMIP® POCIS Glyphosate

Transitions for glyphosate - AMPA and Glufosinate used in water

Analyte	Q1	Q3	Time (ms)	DP (V)	EP (V)	CE (V)	CXP (V)
Glyphosate 1	168	149.8	100	-50	-10	-16	-9
Glyphosate 2	168	62.9	100	-50	-10	-32	-7
AMPA 1	110.1	62.8	100	-50	-10	-24	-9
AMPA 2	110.1	78.8	100	-50	-10	-34	-11
Glufosinate 1	179.9	63.1	100	-50	-10	-58	-9
Glufosinate 2	179.9	135.8	100	-50	-10	-24	-9

Table transitions for glyphosate - AMPA and Glufosinate used for complex matrices

Analyte	Q1	Q3	Time (ms)	DP (V)	EP (V)	CE (V)	CXP (V)
Glyphosate 1	168	62.9	100	-50	-10	-32	-7
Glyphosate 2	168	78.9	100	-50	-10	-50	-3
AMPA 1	110.1	62.8	100	-50	-10	-24	-9
AMPA 2	110.1	78.8	100	-50	-10	-34	-11
Glufosinate 1	179.9	63.1	100	-50	-10	-58	-9
Glufosinate 2	179.9	95.0	100	-50	-10	-24	-5

HPLC Method with LC – MS/MS

Analysis by HPLC – MS/MS (QTRAP 4000)

Column: Acclaim trinity Q1 100mm x 3mm ID (3µm)

Flow rate: 0,5mL/min

Injection volume: 20µL.

Mobile phase: gradient with Ammonium formiate 50 mM pH = 2.9 (A) – Acetonitrile (B)

Time (min)	% A	% B
0	100	0
3	100	0
3,2	0	100
6,0	0	100
6,2	100	0
10,2	100	0