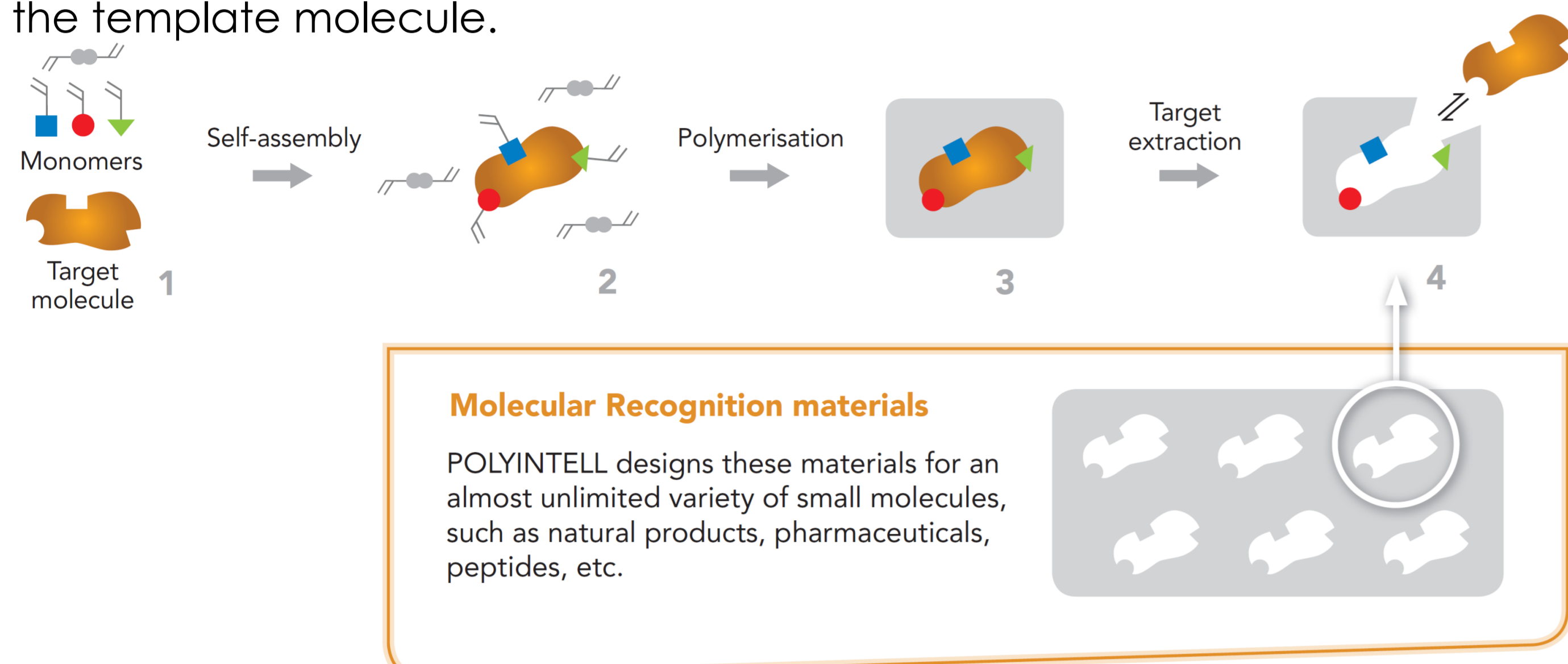


The development of liquid chromatography-mass spectrometry (LC-MS/MS) methods for the simultaneous detection and quantification of a broad spectrum of molecules has made easier the screening of a larger number of samples. In this study, we have developed a unique solid phase extraction (SPE) based on Molecularly Imprinted Polymers (MIP) for multimycotoxins analysis according to regulations. MIPs are polymers with high affinity for some target molecules and good stability to aqueous or organic solvents as well as temperature.

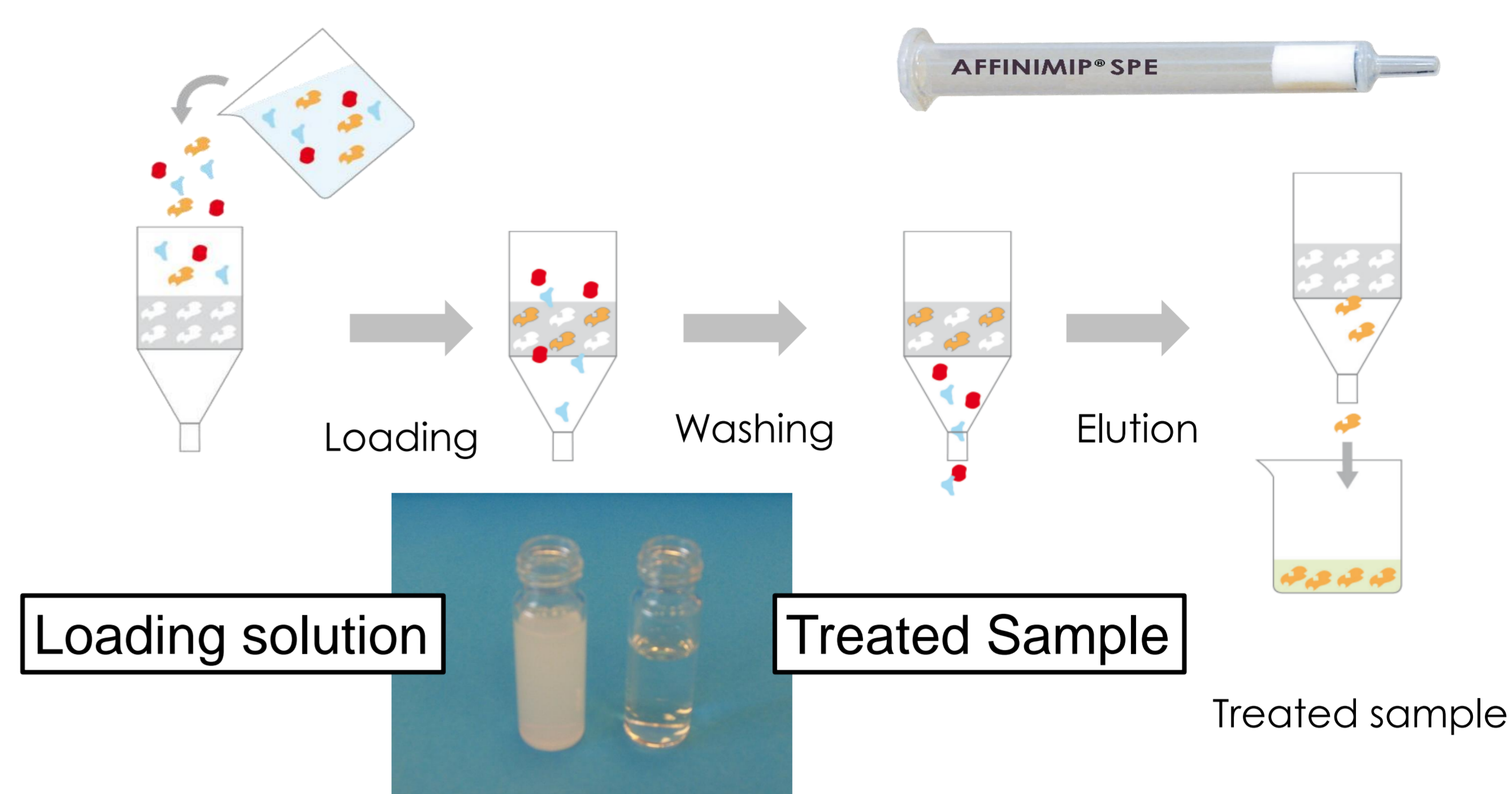
This study shows an efficient clean-up method for a broad family of mycotoxins, **Deoxynivalenol (and its derivatives), Aflatoxins, Ochratoxin A, Fumonisin, Zearalenone, HT-2 and T2 toxins** prior LC-MS/MS detection. By this fast, robust and effective clean-up process, this method enables the analysis of multimycotoxins on cereals and different matrices according to regulations with high recovery yields.

Principle of AFFINIMIP® SPE

Based on molecularly imprinted polymers, AFFINIMIP® is a three-dimensional network that has a « memory » of the shape and functional group positions of the template molecule.



Application of AFFINIMIP® SPE



AFFINIMIP® SPE Multimyco10 3mL: Maize

Extraction of **main regulated mycotoxins** during a **single clean-up** before their simultaneous analyses by LC-MS/MS.

Step	Solvent
Loading	3mL of Cereals extract (25g-100mL 50/50 ACN/Water + dilution 1/5 with water)
Washing	3mL Water 3mL 85/15 Water/ACN Dry 30 seconds
Elution	3mL 48.5/48.5/3 ACN/MeOH/HCOOH

Analyte	C° µg/kg	Mean µg/kg	R%
Σ Aflatoxins	20	15.6	80
Σ Fumonisins	1250	1425	110
Σ HT-2 + T-2	200	150	75
Ochratoxin A	14	12.9	92
Zearalenone	50	50.5	101

Analysis with LC-MS8040 with electrospray source ; Column : Phenomenex Kinetix XB-C18 column ; The quantification was done in MRM mode.

AFFINIMIP® SPE Multimyco LCMSMS (including DON) : Maize

Step	Solvent
Loading	6mL of Cereals extract (25g-100mL 50/50 ACN/Water + dilution 1/10 with water)
Washing	6mL Water-1%acetic acid 2mL 95/5 Water/ACN Dry 3 minutes
Elution	3mL 48.5/48.5/3 Ethyle acetate/MeOH/HCOOH

Analysis by LC-MS
Column : Thermo Hypersil Gold column 50 X 2.1mm id (1.9µm)
Mobile Phase : water-0.1% formic acid/ACN (73/27, v/v)

Analyte	R% (n=2)
Aflatoxin B1	85
Fumonisin B1	96
Fumonisin B2	110
HT-2	86
T-2	96
Zearalenone	96
Deoxynivalenol	82
Ochratoxin A	93

AFFINIMIP® SPE FumoZON

In maize based-babyfood

Step	Solvent
Loading	6mL of extract (25g-100mL 25/25/50 ACN/MeOH/Water + dilution ½ with water)
Washing	6mL 60/40 Water / ACN Dry 3minutes
Elution	2mL MeOH-2% Acetic acid

Sample	C° µg/kg	Mean µg/kg	Recoveries %	% RSD _R
Zearalenone	20	16.9	84.4 (n=4)	1.6
Fumonisin B1	200	168.6	84.3 (n=3)	1.4
Fumonisin B2	200	185.6	92.8 (n=3)	1.9

In maize flour

Sample	C° µg/kg	Mean µg/kg	Recoveries %	% RSD _R
Zearalenone	38	39.2	103.2 (n=8)	8.5
Fumonisin B1	2408	2002.2	83.1 (n=8)	10.3
Fumonisin B1	400	357.6	100.2 (n=2)	-
Fumonisin B2	630	684.6	108.7 (n=3)	11.5

Analysis by LC-MS
Column : Thermo Hypersil Gold column 50 X 2.1mm id (1.9µm)
Mobile Phase : water-0.1% formic acid/ACN (73/27, v/v)

Conclusion

The detection of mycotoxins is a real interest to determine the quality of agricultural commodities. Different AFFINIMIP® SPE have been developed for extraction of mycotoxins and more than 75% recovery were obtained with an efficient clean up (suitable chromatograms with limited interferences).