**Reduction of Matrix Effects of Environmental Waters with Online Solid Phase Extraction LC-MS/MS**

Sonja Krieger, Bettina Schuhn, Agilent Technologies Waldbronn, Germany

**Introduction**

The use of organic compounds leads to contamination of water resources worldwide. Therefore, monitoring these compounds is important. But due to including matrix compounds, a reliable quantification is critical. For online solid-phase extraction (SPE), matrix effects are reduced because the matrix is not retained on the SPE cartridge.

In this work, tap, ground, river, and ultrapure water were spiked with pesticides and analysed with a fast and automated online SPE method coupled to a triple quadrupole mass spectrometry system. To illustrate the reduction of matrix effects, water was spiked with a suite of pesticides (75 ng/L). To determine the spiked concentration and the reduction of matrix effects with online SPE, a standard addition and an external calibration were prepared. By comparing the results of the two calibration approaches, it was observed that the reduction of matrix effects with online SPE is sufficient. Thus, there is no need for a time-consuming standard addition calibration because an external calibration is reliable to quantify compounds in environmental water.

**Experimental**

**System configuration**

The Agilent 1200 Infinity Flexible Cube is the heart of the Agilent 1200 Infinity Series Online SPE solution. The 1200 Infinity Flexible Cube hosts two 2-position/10-port valves for enabling fast method change between direct injection and online SPE. Two trapping columns are alternately in use for time saving and higher sample throughput. The piston pump and solvent selection valve can draw up to three different solvents, and are responsible for loading, cleaning and reconditioning of the cartridges.

With the loading process, matrix components not retained on the cartridges are flushed to waste. The PLRP-S (cross-linked styrene divinylbenzene polymer) cartridges have a long life time, even with samples with high matrix content (> 200 injections). PLRP-S is a highly homogeneous material that is free from silicon and heavy metal ions, and is perfectly suited for the enrichment of medium and nonpolar pesticides.

**Instrumentsation**

All experiments were carried out on an Agilent 1200 Infinity Series Online SPE system comprising:
- Agilent 1200 Infinity Flexible Cube (G4227A), equipped with:
  - Online SPE Starter Kit (G4474A), including one 2-position/10-port valve, 600 bar, capillaries, cartridge holder and cartridges
  - Online SPE Direct Injection Kit (G4474A), including one 2-position/10-port valve, 600 bar, and capillaries
- Agilent 1200 Infinity Binary Pump (G1312B) with LAN card (G1318C)
- Agilent 1200 Infinity Standard Autosampler (G1329B) with 900 µL head (G13130-60002)
- Agilent 1200 Infinity Thermostat Column Compartment (G1315A)
- Agilent 6150 Trise Quadrupole LC/MS System (G6460A) with Agilent Jet Stream technology

**Software**

Agilent MassHunter Workstation Software - Data Acquisition for Quadropole Quadrupole Mass Spectrometer, version B.07.00

**Samples**

Tap water was taken from Waldbronn, Germany, surface water from the Rhein river, and groundwater from Karlsruhe, Germany. The samples and standards were stored at 5 °C, and were centrifuged for 5 minutes at 5,000 rpm prior to injection.

The water samples were spiked with different concentrations of pesticides by diluting a stock solution of 100 µg/L. A five-level calibration curve (10, 25, 50, 100, and 200 ng/L) was prepared in ultrapure, tap, ground, and river water. A standard addition calibration was prepared in river water (75 ng/L) spike plus additionally 18, 25, 50, 100, and 200 ng/L.

**Results and Discussion**

**Performance data of the online SPE method**

Table 1 shows the performance data for the online SPE method like linearity, area precision for two alternating used cartridges and retention time RSD. Additionally, recovery of the enrichment process was determined by comparing peak areas of a 50 ng/L (\( g_0 \)) spike in ultrapure water with direct injection to the same amount of analyte obtained in online SPE mode. Generally, a recovery between 70 to 130 % was considered as acceptable, and all measured compounds are within this range.

**Determination of matrix effects with online SPE and direct injection**

As the recovery for the enrichment process was confirmed (Table 1), it is important to quantify also matrix effects when using LC/MS with electrospray ionization. Therefore, a calibration in ultrapure water and a matrix matched calibration in river and ground water were prepared and the slopes of both calibration curves were compared. It was assumed that in ultrapure water no matrix effects were present. Matrix effects were observed for both environmental (river and ground water) and for most compounds, which is illustrated by comparing matrix matched standards to a calibration in ultrapure water. The analysis of pesticides in ground and river water was performed with direct injection and online SPE (same sample and same method) to compare and determine the matrix effects. Figure 2 and Figure 3 illustrate the example of amincarb. An external enrichment curve (blue) was prepared in ultrapure water and compared to a matrix matched calibration in ground (green curve) and river water (red curve).

**For amincarb, the matrix effects led to overall ion suppression in river water of 27 % for direct injection and just 16 % for online SPE. For ground water direct injection showed 22 % suppression and online SPE 7 % ion enhancement. The other compounds, which are not shown here, also indicate the same effects with more or less intensity. For most compounds, less matrix effects were detected with online SPE measurements compared to direct injections.**

Amincarb is an interesting example that shows different matrix effects with direct injection and online SPE. While direct injection leads to ion suppression, online SPE showed negligible ion enhancement. This shows that the enrichment process parts of the matrix, such as salts and minerals, are flushed out and matrix components cannot interfere with the analyte.

**Standard addition calibration and external calibration**

When matrix effects are sample dependent, it is recommended to use the standard addition calibration (SC). For the following experiments, river water was spiked with 75 ng/L pesticide standard. Additionally, for standard addition calibration five aliquots of increasing standard concentration were added (10, 25, 50, 100, and 200 ng/L).

For comparison, an external calibration (EC) was prepared in drinking water, as an EC is less time and labor consuming, especially when many samples have to be measured. Finally, the results of EC and SC calibration were compared and the spiked samples were evaluated. The measured concentration of EC and SC is shown in Figure 4 as well as a ~ 20 % tolerance of 75 ng/L spike. Figure 4 shows the results for external calibration and standard addition calibration for online SPE.

In summary, the external and standard addition calibration achieved good results and precision for the determination of 75 ng/L spiked in river water. The results were comparable for both calibration methods, which indicate that only SPE online can be used as calibration is perfectly suited for the quantification of organic compounds in environmental water and the standard addition approach is not mandatory.

**Conclusions**

- This work demonstrates the reduction of matrix through the enrichment process of the online SPE. During the loading procedure, different solvents can be selected to wash matrix to the waste.
- With online SPE, standard addition calibration is not necessary, and an external calibration is sufficient to quantify organic compounds in environmental water, which saves time and labor.
- No carryover was determined, which indicates a successful cleaning procedure of the cartridges with the Agilent 1200 Infinity Flexible Cube.
- The recovery of the online SPE analysis was evaluated and showed good results as well as linearity, area and retention time precision.

**References**

Sifahm B. Reduction of Matrix Effects with the Agilent 1200 Infinity Series Online SPE Solution, Agilent Technologies Application Note, publication number 5991-5346EN

![Figure 1: Agilent 1200 Infinity Series Online SPE Solution coupled to an Agilent 6410 Triple Quadrupole Mass Spectrometer](image1)

![Figure 3: Matrix effect for amincarb in river water (red, 16 % ion suppression) and ground water (green, 7 % ion enhancement) calibrated in ultrapure water (blue) and measured with online SPE.](image3)

![Figure 4: Comparison of the standard addition calibration to an external calibration in drinking water. A 75 ng/L amount of pesticides were spiked in river water.](image4)