PESTICIDE RESIDUE RESEARCH GROUP

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Application of microLC-MS/MS in pesticide residue determination in fruits, vegetables and fruit jams

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INTRODUCTION

LC high flow rates such as 0.5 – 1 mL/min theoretically can facilitate and increase the signal response of the target compounds when it is coupled to a mass spectrometric detector. But, the flow rate influence on the mass spectrometer response is very dependent of the interface characteristics and matrix effects as it is observed in many cases. In some ESI designs a decrease to below 0.1 mL/min can be optimum as a consequence of an increase in ion production and sampling. Microsprays with small plumes and low droplet space charge repulsion coupled with adequate ESI source design represents two key aspects in an increasing ion production and sampling. In this work we have explored the capabilities of micro flow LC in improving MS signal response and decreasing matrix effects.

Fruits and vegetables are complex matrices and it could lead in strong matrix effects. Various strategies can be used with the aim to minimize or eliminate the matrix interferences. Dilution is an easy and effective method to get rid of interfering compounds, obtaining this way the injection of less matrix load into the chromatographic system, although in this case sensitivity is a key factor, given that it implies a reduction in the amount of analytes but the high sensitivity of microLC-MS systems makes possible application of very high dilution factors.

QuEChERS extracts of 4 blank matrices (tomato, green pepper, orange and grape jam) were spiked at eight concentration levels with 90 pesticides and at seven levels with 107 pesticides (jam), diluted 30 times and analysed by microLC-MS/MS. For separation, a C_{18} narrow-bore column was used (0.5 mm x 50 mm x 2.7 µm). Data were used to evaluate linearity (r^2), reporting limits, LOQs, matrix effects (%), precision (RSD%), repeatability and reproducibility (n=5) at two different concentrations. An application of the method was carried out analyzing 61 fruits and vegetables samples of different commodity groups and 51 jam samples .

EXPERIMENTAL

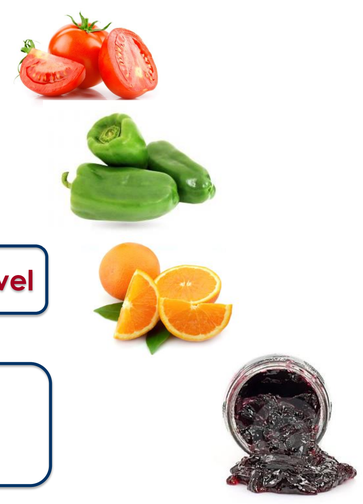
Sample handling

Citrate buffered QuEChERS

Blank extracts

Spiking at appropriate level

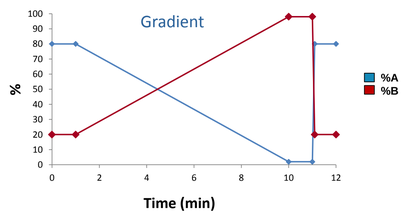
Diluted 30 times with AcN:H_2O (20:80)



microLC-MS/MS

System: Ekspert microLC 200 coupled to an 4500 QTRAP ABSciex

HPLC parameters:
- Injection volume: 5µL and 3 µl (jams)
- Flow rate: 30µL/min
- Column: HALO C18 2.7 µm 90 Å 0.5x50mm
- Mobile Phases and gradient :
  - A → H_2O 0.1% formic acid
  - B → AcN 0.1% formic acid

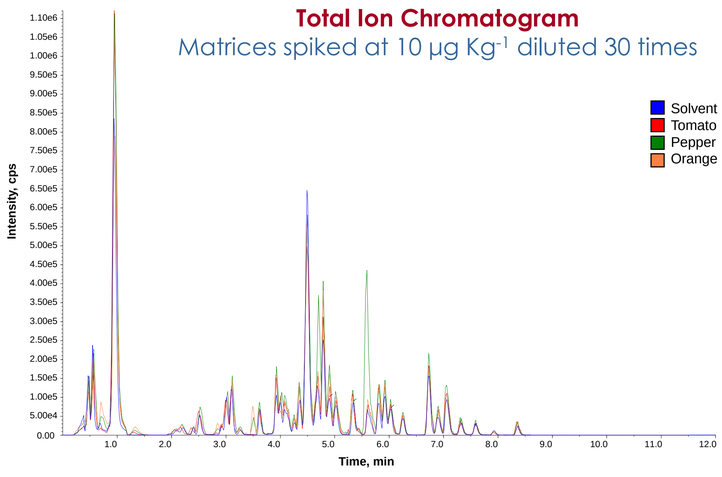


Ekspert microLC 200

MS parameters:
- Ion source: ESI with microFlow electrode
- Polarity: Positive and Negative
- Schedule MRM software features

ABSciex 4500 QTRAP

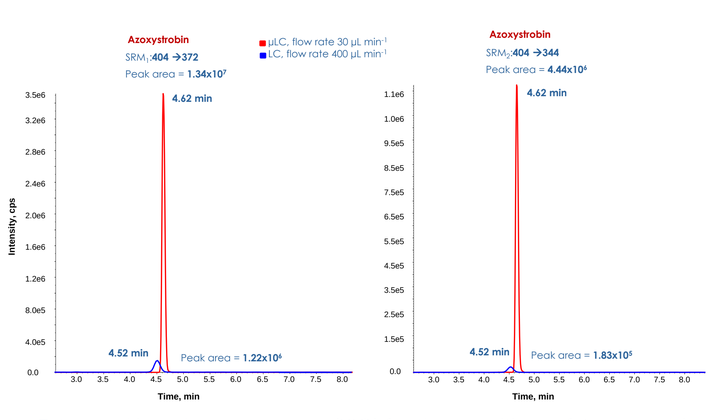
RESULTS



Total Ion Chromatogram
Matrices spiked at 10 µg Kg^{-1} diluted 30 times

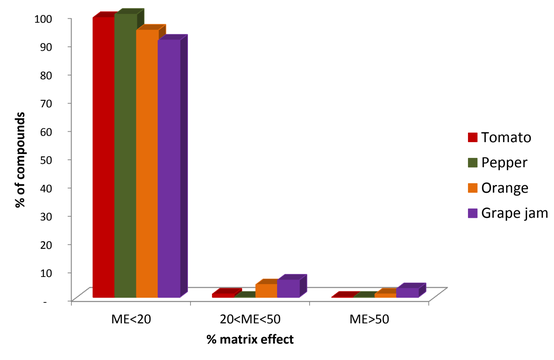
Total ion chromatograms of a tomato, pepper and orange extracts spiked at 10 µgm Kg^{-1} and diluted 30 times.

High sensitivity gain by microflow-LC-ESI-QqQ-MS

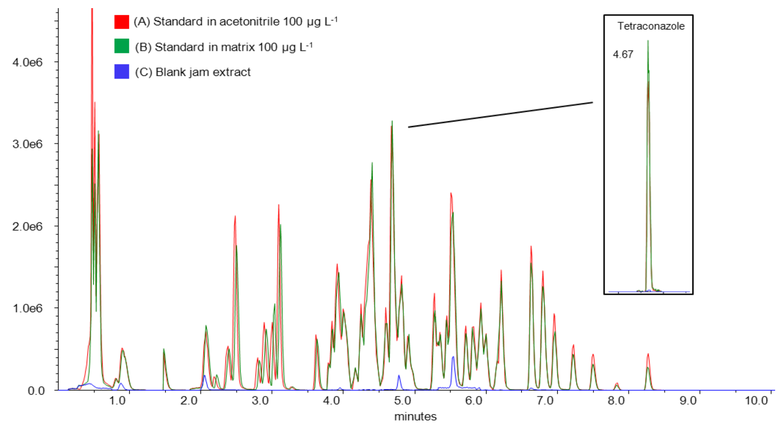


Extracted ion chromatograms (XIC) for the transitions of Azoxystrobin corresponding to a spiked orange matrix at a concentration of 10 µg Kg^{-1} comparing a conventional LC with a microLC

Matrix effects

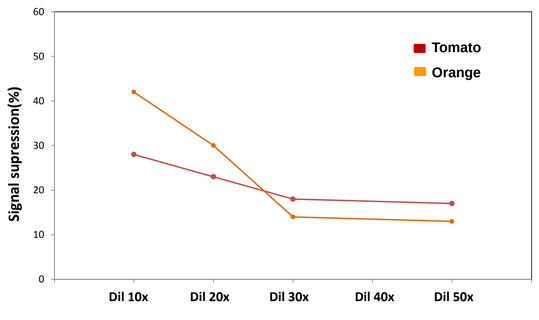


Matrix effects comparison of the four studied matrices (tomato, pepper, orange and grape jam) diluted 30 times.



Total ion chromatograms of a standard pesticide mixture solution in acetonitrile at 100 µg L^{-1}, standard pesticide mixture solution in blank grape jam extract at 100 µg L^{-1} (150 µg kg^{-1}) and blank grape jam extract.

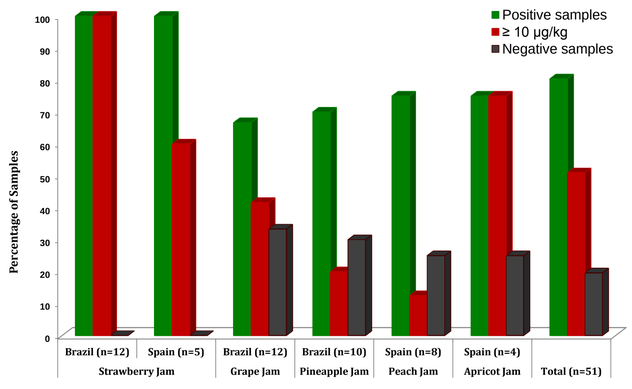
Influence of dilution factor



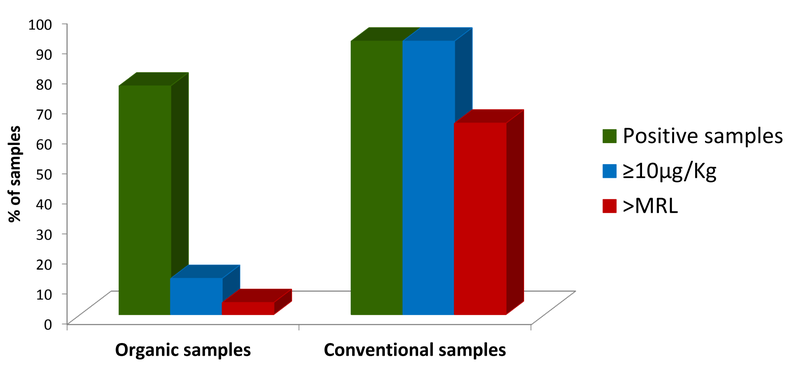
Influence of dilution factor on matrix effect. An example with Dicrotophos, in tomato and orange matrices.

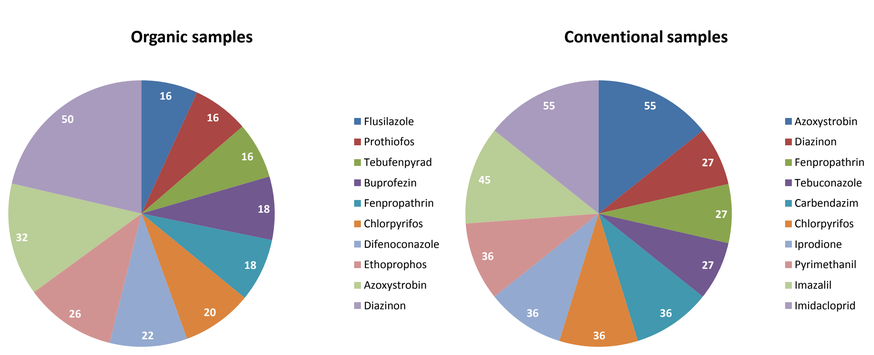
Application of the method

Fruit Jam samples

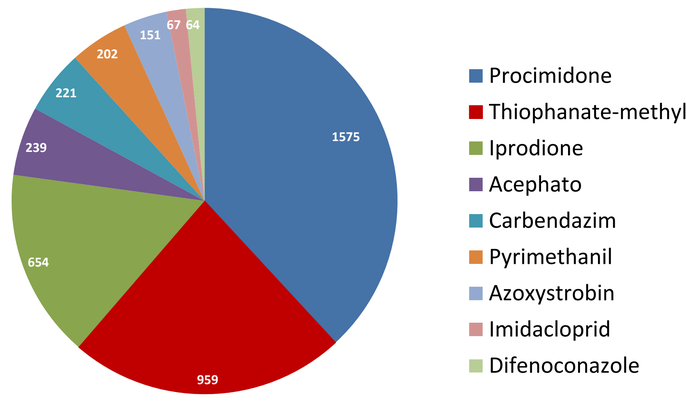


Fruit and vegetable samples





Pesticides more frequently detected in organic and conventional samples



Pesticides detected al highest concentrations (µg/kg)

CONCLUSIONS

- The method was validated studying recoveries, reporting limits, linearity , repeatability and matrix effects, giving good results.
- Sensitivity of the system was enough to determine the majority of the pesticides spiked at the lowest level (5 µg/Kg) with reporting limits corresponding to a signal-to-noise ratio higher than 20.
- The commercial microLC pump and narrow bore columns used, assured good retention time as well as peak area reproducibility.
- It was applied to samples to 61 fruits and vegetables samples of different commodity groups, 82% were organic and 18% conventional samples. The percentage of positive samples in the organic and conventional group of samples was 76 and 91%, respectively. In total, were detected 41 pesticides. The method was also applied to 51 jam samples. In total, were detected 42 pesticides, 80% of samples were positives for at least one pesticide.
- The microflow-LC-ESI-QqQ-MS method developed can provide better sensitivity than methods based on conventional flow rates and allow sample dilution of up to 30-fold, thus minimizing this potential for matrix effects.

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