The determination of Thiram residues in fruit by UPLC-MS/MS

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Introduction
- Thiram is a non-systemic dimethyl dithiocarbamate fungicide, mainly used in the field as well as to protect harvested crops during transport and storage.
- Considered to be a general use pesticide, licensed for use on a range of crops including fruit, vegetables and ornamentals to prevent fungal diseases.
- Also used as an animal repellent to prevent damage caused by rabbits, rodents and deer.
- The analysis of dithiocarbamates is generally performed by the measurement of liberated CS₂, following decomposition in the presence of SnCl₂/HCl.
- This technique does not distinguish between the other related compounds that also produce CS₂ in this way.
- There was a requirement to develop and validate a modern analytical method that was specific to Thiram.
- This method had to be suitable for routine use for the analysis of samples generated in crop residue trials.

Challenges
- Thiram can undergo decomposition when exposed to acidic plant juices.
- It is also suggested that Thiram stability is adversely affected by the presence of copper ions.
- A fast efficient LC-MS method was required that could complement the current CS₂ screening technique.

Summary of extraction and clean-up procedure
- Frozen crop samples were prepared for analysis by homogenisation with dry-ice.
- Direct Thiram analysis in fruit (strawberry and apple) involved extraction by shaking with cold acetonitrile in the presence of anhydrous sodium sulfate.
- An aliquot was diluted with an aqueous ETDA solution prior to quantification by UPLC-MS/MS.
- No further sample clean-up stage is required as there were no observable matrix effects using this approach.
- This method allows a range of fruit sample types to be analysed, using this quick, efficient, robust and reliable technique.
- The analytical method was developed to utilise UPLC-MS/MS which significantly reduced the analysis time to approximately four minutes per sample injection.
- Two MS/MS ion transitions can be simultaneously monitored to demonstrate a suitable confirmatory technique.
- In addition, the use of the alternative CS₂ technique can offer another complimentary technique if used alongside the Thiram specific LC-MS/MS method.

The MS/MS scan of m/z 241 showed fragmentation of the molecular ion to produce daughter ions at m/z 120 and m/z 88.

Conclusions
- It is possible to analyse Thiram in fruit matrices using UPLC-MS/MS.
- This gives rise to short analysis times, enabling a large number of samples to be quantified in a single batch.
- The use of UPLC-MS/MS means that extraction and clean-up procedures can be simplified due to the high instrument selectivity obtained.
- This technique offers an improvement over the CS₂ approach being specific to Thiram and avoiding possible false positive results.
- The analytical method has been validated on strawberries and apples at two concentration levels with five replicates per level.
- The method was demonstrated to be robust with recovery values falling within the range 73-117%.
- The overall mean recoveries between 89-97 % and coefficient of variation values of <13% show that this method is suitable for routine sample analysis.
- Additional confirmatory data can be acquired using a second MS/MS transition, as well as obtaining further information by performing additional analyses using the CS₂ technique.

References