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Note d'application

# Evaluation of the Potential of the ACQUITY QDa Mass Detector for Use in Forensic Chemistry and Drug Control Laboratories

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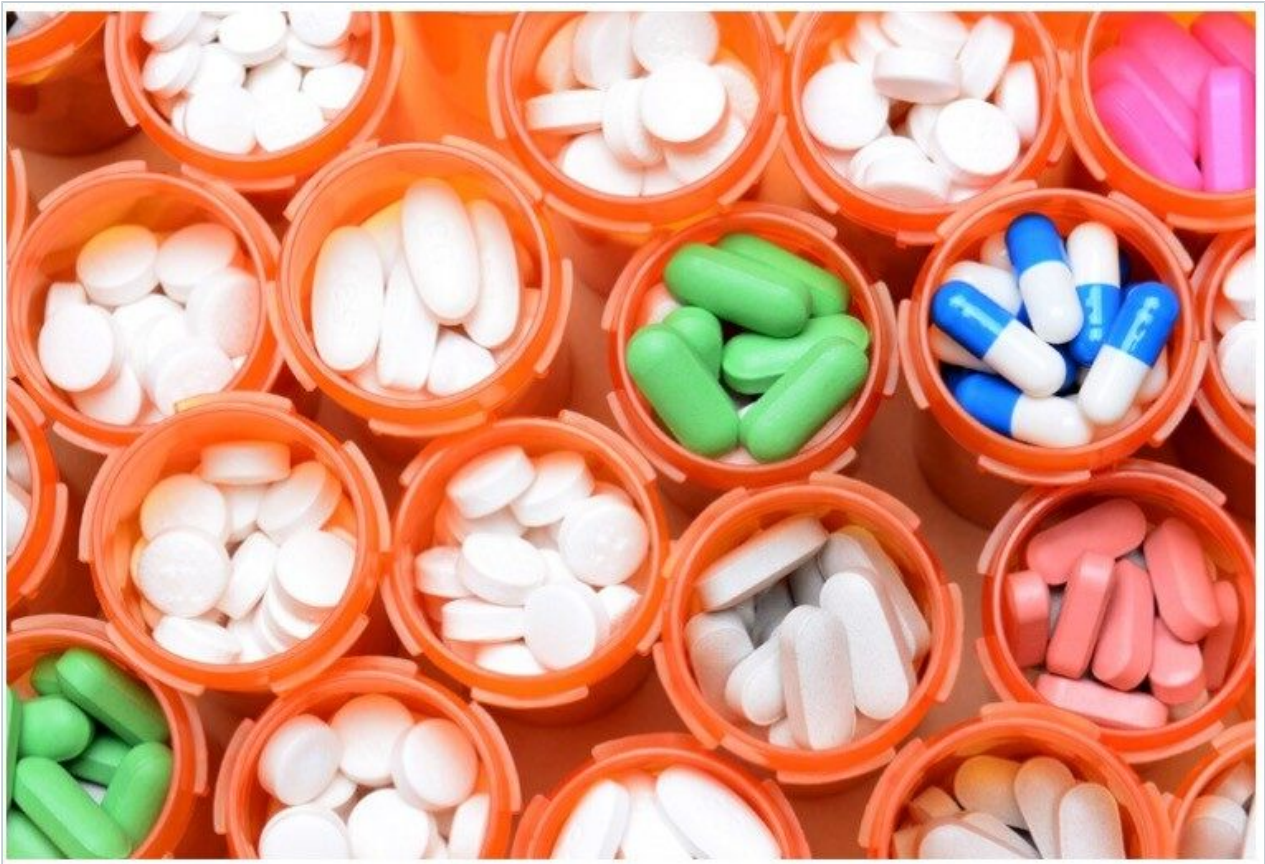

*For forensic toxicology use only.*

This is an Application Brief and does not contain a detailed

Experimental section.

## Abstract

This application brief assess the transferability of an existing toxicology library to the Acquity QDa mass detector.

### Benefits

Application of an existing toxicology library to the qualitative screening of medicines using the ACQUITY QDa — a promising tool for drug control.

## Introduction

In recent years, a comprehensive spectral library for use in analytical toxicology has been developed. The library was originally generated using the Waters ACQUITY TQD Mass Spectrometer and was prepared by acquiring full scan mass spectra over multiple cone voltages, to yield compound-specific fragmentation patterns by the process of in-source collision-induced dissociation.[1] Since the first application of this methodology, over a decade ago, the approach has been applied to newer generation instruments[2] and the library has been expanded; it now contains data for over 950 toxicologically-relevant substances. The purpose of the current work was to evaluate the feasibility of using the existing library in combination with the ACQUITY QDa[3] to provide a simple, low-cost, qualitative screening and identification system for use in forensic chemistry and drug control laboratories. For this study, a selection of over-the-counter and prescribed medicines were analyzed as representative agents.


*The ACQUITY UPLC I-Class System and ACQUITY QDa Mass Detector.*

## Experimental

### Materials

Preliminary spectral testing was performed for a range of licit and illicit drug substances, using mixtures prepared from certified reference  material (Sigma-Aldrich). Eight mixtures, each containing ten compounds, were analyzed.

Authentic samples for subsequent testing were prepared using a selection of over-the-counter and

prescribed medicines in tablet, capsule, or liquid form.

### Sample preparation

Individual tablets/capsules or 250 µL of medicines supplied in liquid form, were added to 25 mL of a methanol and water mixture (70:30) and sonicated at room temperature for 30 minutes. One millilitre of the resulting solution was transferred to a 2 mL microcentrifuge tube and centrifuged at 13000 rpm for 5 minutes. Fifty microlitres of the supernatant was diluted with 950 µL of water in a maximum recovery vial and vortex-mixed. LC-MS analysis was performed using 10 µL of the resulting solution.

### LC-MS method conditions

Chromatographic separation was achieved within 15 minutes using an ACQUITY UPLC I-Class (FTN) and an established toxicology screening gradient.[1,2,4] The ACQUITY QDa was operated in ESI+, and full scan data were acquired over a *m/z* range of 80–650 at the following five cone voltages: 10 V, 20 V, 35 V, 45 V, and 55 V.

## Results and Discussion

Data for a selection of medicines were acquired using the ACQUITY UPLC I-Class System combined with the ACQUITY QDa according to a well-established technique in which samples are screened against a library comprising reference retention time and multiple spectra.[1,2,4]

A preliminary assessment of voltages was performed using mixtures of drug standards at a concentration of 200 ng/mL. Data were acquired using the ACQUITY QDa at identical voltages to those used in the preparation of the original ACQUITY TQD library, i.e. from 20 V to 95 V in increments of 15 V. This initial data indicated that for the same cone voltages, the ACQUITY QDa exhibited increased fragmentation; consequently library voltages were adjusted to achieve parity with the ACQUITY QDa. The modified library was subsequently applied to the analysis of eight pharmaceuticals.

The multi cone voltage data acquired for each of the eight samples were processed using MassLynx Software with ChromaLynx Application Manager which detects the components within each sample and provides an identification through library matching. The confidence with which a substance is identified is presented as an average library match factor which has a maximum value of 1000. The average match factor is determined by comparing the measured and library spectra acquired over the five cone voltages.

The active ingredients detected in each of the eight samples are listed in Table 1 together with the average match factors as determined by ChromaLynx. For one of the medicines, Imuran, there was no match with the library, however, a large response was observed at the same retention time for each of the five cone voltages. The package insert for the product indicated that the active ingredient in Imuran is azathioprine, and this was consistent with the spectral data shown in Figure 1. The acquired data was subsequently used to generate library entries for azathioprine.

| Medicine | Active ingredients detected (match factor) |
|---|---|
| LEMSIP® Max Cold and Flu Remedy | Phenylephrine 6.1 mg (707), paracetamol 500 mg (804), caffeine 25 mg (897) |
| Galpharm™ Hayfever and Allergy Relief | Cetirizine 10 mg (832) |
| Entrolax® | Bisacodyl 5 mg (889) |
| Galpharm Extra Power Pain Relief | Paracetamol 200 mg (816), caffeine 45 mg (880) |
| Buscopan® | Scopolamine butylbromide 10 mg (902) |
| Prozac® | Fluoxetine 20 mg (883) |
| Benylin® | Guaifenesin 100 mg/5 mL (701) |
| Imuran® | Azathioprine 50 mg* |

*Table 1. The active ingredients in each of the medicines analyzed, together with the disclosed amount and the average match factors to the library entries.*

**Azathioprine was not present in the library and the measured spectra were used to create a library entry.*

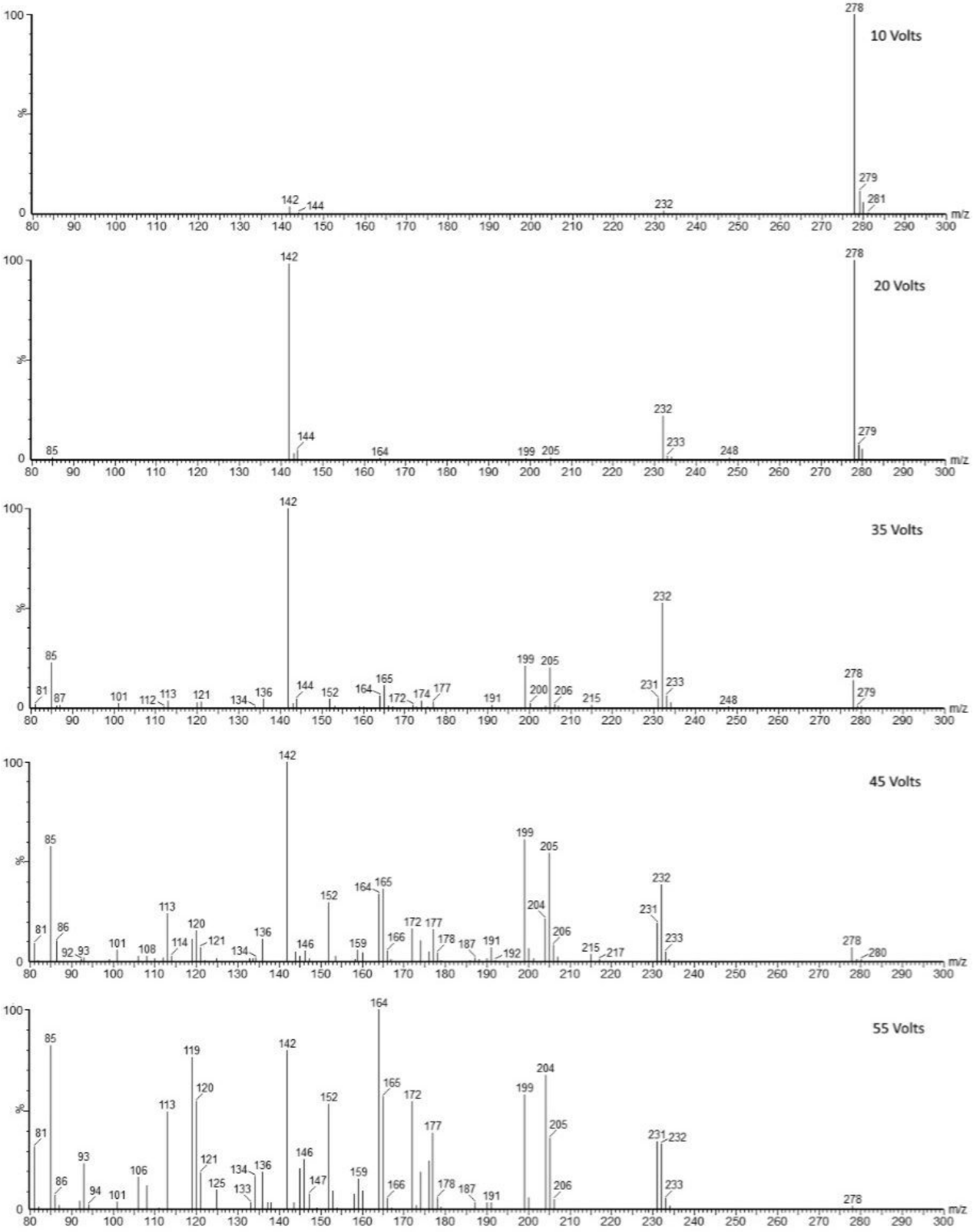


*Figure 1. ESI+ spectra for azathioprine (precursor mass m/z 278) on the ACQUITY QDa at the following cone*

*voltages: 10V, 20V, 35V, 45V, and 55V.*

An example of the information available in the browser of the ChromaLynx Application Manager is shown in Figure 2 for the analysis of the LEMSIP MAX Cold and Flu Remedy.

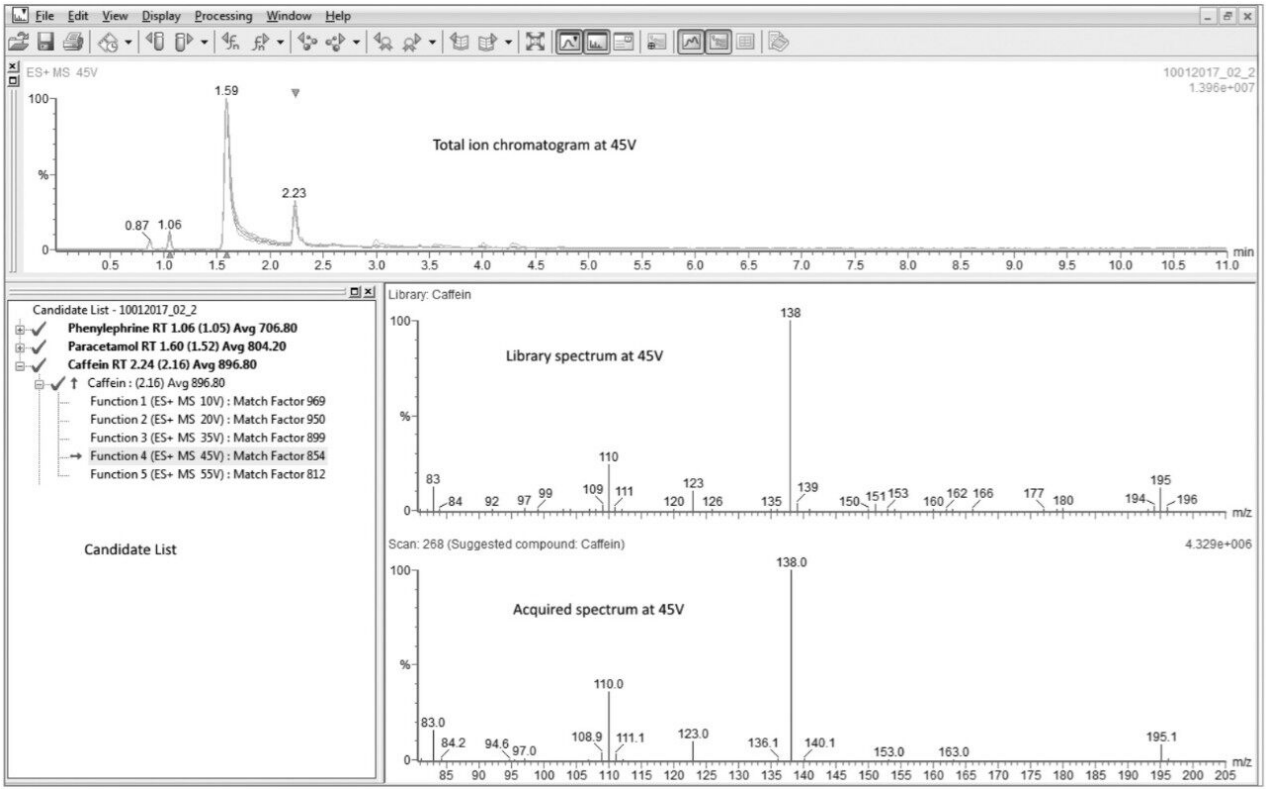


*Figure 2. The ChromaLynx browser displaying the results of the analysis of the LEMSIP Max Cold and Flu Remedy, highlighting the identification of caffeine with a precursor ion at m/z 195.*

## Conclusion

In this study, a series of representative medicines were used to assess the feasibility of applying an existing toxicology library to the ACQUITY QDa Mass Detector. Application of an established chromatographic method together with the qualitative screen demonstrated very good agreement between library spectra and acquired data, leading to the identification of the active ingredients in the medicines. Therefore, the modified

toxicology library, in combination with the ACQUITY QDa, appears promising as a low-cost solution for forensic chemistry and drug control testing.

## References


1. Screening of Xenobiotics by Ultra-Performance Liquid Chromatography-Mass Spectrometry Using In-Source Fragmentation at Increasing Cone Voltages: Library Constitution and an Evaluation of Spectral Stability, L. Humbert, F. Grisel, C. Richeval and M. Lhermitte, *Journal of Analytical Toxicology* 2010; 34: 571–580.
2. Systematic Toxicological Screening Using the ACQUITY UPLC I-Class/Xevo TQ-S micro, R. Lee and M. Wood, 720005661EN, 2016.
3. ACQUITY QDa Detector Brochure. 720004632EN.
4. Postmortem Drug Screening by Non-Targeted and Targeted Ultra-Performance Liquid Chromatography-Mass Spectrometry Technology, T. Rosano, M. Wood, and T. Swift, *Journal of Analytical Toxicology* 2011; 35: 411–423.


## Featured Products

ACQUITY UPLC I-Class PLUS System <https://www.waters.com/134613317>

ACQUITY QDa Mass Detector <https://www.waters.com/134761404>

720006004, May 2017