



Direct TLC-MS analysis using the 4000MiD®

Microsaic Systems plc, GMS House, Boundary Road, Woking, Surrey, GU21 5BX, United Kingdom

INTRODUCTION

TLC-MS is growing in popularity, with applications including identification and elucidation of unknown substances in research, forensic and environmental fields. TLC-MS system provides an easy way to elute a 4mm diameter zone and direct it to the MS to obtain a mass spectrum of the substance adsorbed on the stationary phase at that location. Integration of the TLC-MS interface with the miniaturised, deployable, single quadrupole 4000 MiD® mass detector provides a completely stand-alone and easy to use solution for TLC spot identification. This note describes the simplicity with which both systems can be integrated, and shows examples in spot identification in the areas of synthetic chemistry and natural products.

MICROSAIC 4000MiD® and MiDas™

The 4000MiD® is a self-contained and deployable single quadrupole mass detector with NO external vacuum pumps, integrated PC and 'plug and play' components that allow non-mass spectrometrists to operate and maintain the equipment with minimal effort and training. The MiDas™ is a compact interface module used to enable mass detection for a multitude of applications. It is a separate unit to the MS, fully controlled by the 4000MiD® and associated method files within the on-board Masscape® software.



FIGURE 1. 4000MiD® and MiDas™



FIGURE 2. TLC-MS interface

TLC-MS INTERFACE

The utilities used to run the TLC-MS interface are supplied by the 4000MiD® and MiDas. The interface is pneumatically driven and uses the same gas supply as the 4000 MiD® (2-6 bar gas pressure, in this case 99.5% purity nitrogen).

Elution is achieved using the analytical quality pump present in the MiDas™, and integrates simply, using the 4000MiD® on-board Masscape® software which takes care of data acquisition and provides a simple and intuitive platform for data analysis.

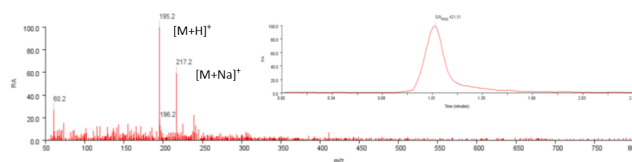


FIGURE 3. Mass spectrum obtained from a 100ng caffeine TLC spot, with the Extracted Ion Chromatogram (EIC) for [M+H]⁺ inset.

SENSITIVITY

TLC-MS of a 100ng caffeine spot was found to give a S/NRMS above 400 using the EIC trace, and a clear caffeine mass spectrum (see Figure 3).

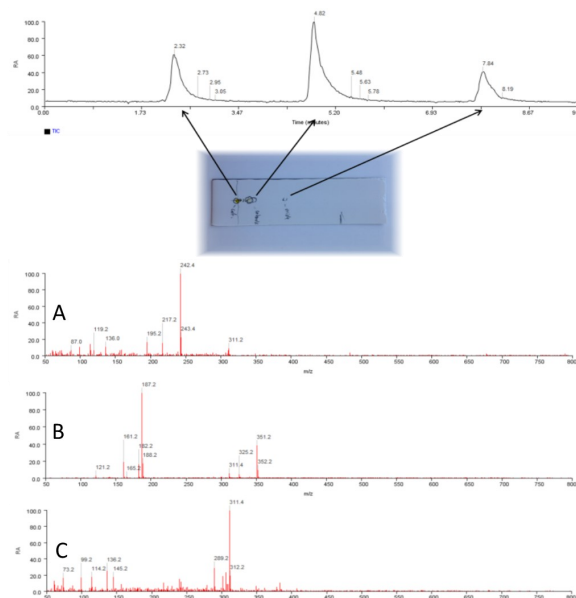


FIGURE 4. TIC and mass spectra of the 3 components from a 'real' TLC plate

NATURAL PRODUCT IDENTIFICATION

Hypericum perforatum, known as St John's wort, is a flowering plant of the genus *Hypericum* and a medicinal herb with antidepressant activity and potent anti-inflammatory properties. The crude preparation of this plant material contains a plethora of chemical compounds, including flavonoids, phenolic acids and saturated fatty acids. Work previously performed by CAMAG has been duplicated in order to separate flavonoid compounds by TLC and then obtain mass spectra from the resulting spots to confirm the identity of these compounds.

EXPERIMENTAL

100mg of a St John's wort formulated product was extracted in 5mL of neat methanol, and 1 μ L used to spot a silica gel 60 F 254 plate. The plate was run using a mobile phase of ethyl acetate - dichloromethane - formic acid - acetic acid - water 100:25:10:10:11 and the resulting fluorescence observed at 370nm

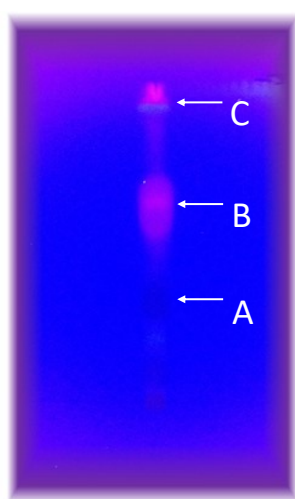


FIGURE 5. Chromatographed TLC plate viewed under 370nm light

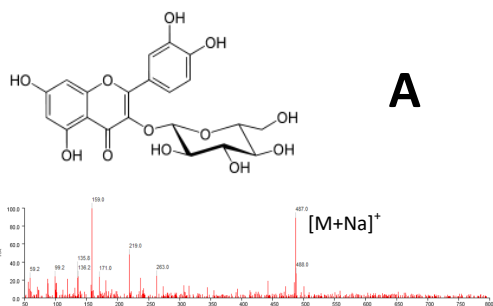


FIGURE 6. Structure and mass spectrum obtained from TLC plate for isoquercetin

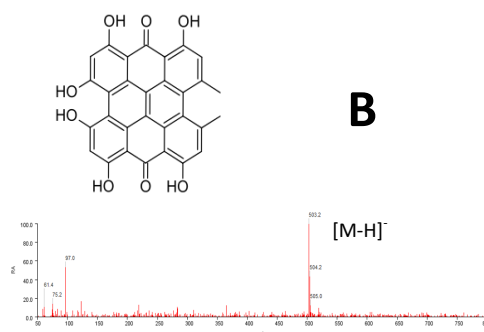


FIGURE 7. Structure and mass spectrum obtained from TLC plate for hypericin

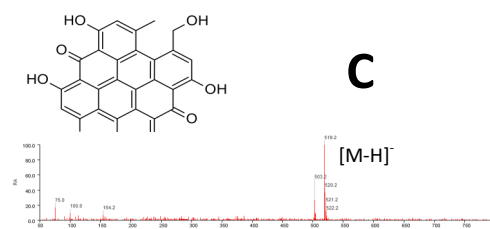


FIGURE 8. Structure and mass spectrum obtained from TLC plate for pseudo-hypericin

RESULTS

The 4000 MiD®, MiDas™ and TLC-MS interface have been shown to work together 'straight out of the box' to produce quality mass spectra from real-life samples. The integrated gas and liquid handling supplied by the 4000 MiD® and MiDas™, controlled by the on-board Masscape® software give a high level of integration that doesn't rely on external pumps and additional gas regulators.

CONCLUSIONS

Microsaic has proven another powerful application that can be achieved using the deployable and stand-alone 4000 MiD® and MiDas™ system, differing from alternative TLC-MS systems that require non-integrated pumps and secondary gas supplies.

The system performed consistently well, with the ambient microspray source that means far less stationary phase related deposits reach the source and diminish performance. Unlike conventional MS systems, the source and interface components are changeable in minutes by non-experts.

The TLC-MS configuration allows for both spot identification and direct injection via the injection port of the MiDas™. The deployable and versatile nature of the 4000 MiD® and MiDas™ means the system does not have to be a dedicated 'TLC-MS', but can also be used in combination for a multitude of applications in your laboratory, as easy as moving a PC.