Multisample Probe for Fast-Atom Bombardment / Mass Spectrometry

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An inexpensive multisample fast-atom bombardment (FAB) probe assembly was designed for high-throughput analysis of samples on a VG ZAB-SE mass spectrometer. The system consists of a vacuum lock system and a FAB probe whose tip contains five or more sample wells. The probe enters the mass spectrometer source region perpendicular to the secondary ion beam axis. The probe is maintained at high voltage on contact with a spring clip attached to the screen plate of the source block. Sample throughput with the multisample probe is twice that of a coaxial probe, with about twice the sensitivity and no sample cross contamination. (*J Am Soc Mass Spectrom* 1993, 4, 504-506)

The traditional method for the analysis of samples by fast-atom bombardment (FAB) is to load a single sample onto a probe target and then insert the probe into the mass spectrometer through a vacuum lock. A number of single-sample FAB analysis methods have been developed for which special probe targets have been designed, including targets for high sample loading of high-mass molecules [1] and for thermally assisted FAB [2, 3]. Rotatable FAB probes are available that can be used for the consecutive analysis of two FAB samples [4, 5]. Two samples can also be analyzed with split and articulated ("wobble") targets [3, 4] but cross contamination of one sample with the other is common. The rotatable FAB probes and split and articulated FAB targets were originally designed for the consecutive analysis of an unknown and reference material in exact mass measurements.

In our laboratory environment, large numbers of FAB samples are analyzed to support pharmaceutical development. An inexpensive and routine method was needed to increase the productivity in generating large numbers of high-quality, low- and high-resolution FAB spectra. Flow-FAB [6] and thin-layer chromatography FAB [7, 8] were evaluated and found not to produce the same quality spectra as by probe analysis. Robotic methods for sample introduction were considered but not pursued because of the complexity and expense to develop. We designed a multisample FAB probe for the consecutive analysis of five (or more) samples. With this probe, FAB spectra are produced with equal or better quality than those with the single-sample

probe but are generated with minimal effort and twice the speed. In addition, the design chosen requires no major modifications to the existing spectrometer system, thereby minimizing implementation costs. The new multisample probe is an extension of the "reference" probe design introduced by VG (Manchester, UK) for flow-FAB [9].

A VG ZAB-SE mass spectrometer equipped with an "open" FAB source, originally designed for flow-FAB, and a vacuum lock perpendicular to the spectrometer beam axis was used. A schematic diagram of the multisample probe and source assembly is illustrated in Figure 1. A rectangular probe tip, 72 mm $\times$ 5 mm $\times$ 1.5 mm, containing five (or more) sample wells was fabricated from stainless steel and polished. A polished surface improves the adhesion of the sample and matrix to the surface and minimizes sample spreading from the confines of the well. The wells are 2.5 mm wide, 1 mm deep, and spaced 5 mm apart. Approximately 1 μ L of dilute matrix and sample are respectively placed at the top of each well after the probe tip is rotated on its axis and held at approximately 45°. The samples flow into the wells, completely filling them. The multisample probe tip is introduced into the FAB source perpendicular to the spectrometer beam axis via the vacuum lock extension. The vacuum lock extension is 3 in. long to accommodate the elongated probe tip for evacuation before introduction to the high vacuum of the spectrometer. "O" rings at both ends of the inlet extension serve to seal the system on insertion of the probe. After evacuation, the high-vacuum valve is opened, and the probe is pushed into the high-vacuum region until constrained by the probe stop. At this point, the probe guide is locked into place, with the thumb screw located on the probe guide extension. The translation of the probe and tip in the source region is controlled by a high-performance Mitutoyo (MTI Corp., Paramus, NJ) vernier dial (model

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¹VG originally marketed a "wobble" probe for the ZAB mass spectrometers and presently sells a probe with two targets for the Autospec mass spectrometer series. These probes are designed for accurate mass measurements.

MULTIPLE TARGET FAB PROBE ASSEMBLY
(EXPLODED VIEW)

Figure 1. Exploded view of the multiple-target FAB probe assembly. The original VG components are the end flange, high-vacuum valve, and FAB source (excluding the spring contact clip).

297-201). As illustrated in Figure 2a, when the probe tip enters the source block it makes electrical contact with a flexible stainless steel clip. This spring clip attaches securely around the screen plate, which floats at the source potential. The clip can be easily disconnected from the screen plate without disassembling the source. (Exploded views of the spring clip and the multiple-target tip are also illustrated in Figure 2b and c, respectively.) Each sample well is fully irradiated by the cesium ion beam. Transmission of the secondary ions is very efficient owing to the closeness of the irradiated samples to the ion optics. No cross contamination of samples is observed.

Sensitivity measurements of Cs_2I^{1+} at m/z 393 by a Faraday detector with the multisample FAB probe (1.1×10^{-11} A) were found to be approximately twice that of the coaxial probe when normalized to the same

MULTIPLE TARGET FAB PROBE ASSEMBLY
(TOP VIEW)

Figure 2. (a) Top view of the multisample FAB probe and source assembly. Exploded views of (b) the stainless steel clip and (c) multiple-well target tip. The tip of the multi-target FAB probe is extended into the FAB source region with a sample well irradiated by the cesium ion beam. The stainless steel spring clip is attached securely to the screen plate elevating the probe tip to the source potential.

target surface areas. Part of this improvement can be accounted for in that the multisample target well is approximately 0.5 mm closer to the source slit created by the ion repeller plates (2.5 mm) versus the target of the coaxial probe. FAB data acquired in the consecutive analyses of five different pharmaceuticals from five adjacent sample wells are illustrated in Figure 3a-e, demonstrating the absence of any cross contamination from adjacent samples.

The multisample probe assembly has been in continuous use now in our laboratory for over nine months. The throughput of low-resolution FAB samples has increased by approximately 50%. In the high-resolution peak-matching mode, pairs of adjacent samples, each with internal standards [10], can be rapidly analyzed without additional tuning. To maximize the number of samples analyzed, one does not use an excessive amount of FAB matrix so as to keep the source pressure as low as possible during the analysis but not too sparingly so that the matrix does not evaporate before the last analysis. For lengthier analyses, this can be achieved by using slightly more matrix for the later samples versus the earlier samples. The less volatile matrixes, *m*-nitrobenzyl alcohol and a 5:1 (w:w) mixture of dithiothreitol:dithioerythritol, have been found to work best with the multisample probe for a wide variety of pharmaceuticals. Vials of the latter matrix mixture were also adulterated with salts so that successive analyses using pure and adulterated

Figure 3. Consecutive spectra obtained with the multisample FAB probe demonstrating the absence of cross contamination between adjacent samples. The consecutive FAB spectra are (a) substance P, fragments 5–11 [molecular weight (MW) 868]; (b) tetracycline (MW 444); (c) platelet-activating factor antagonist (MW 631); (d) piperacillin (MW 517); and (e) erythromycin (MW 733). Matrix 5:1 (w:w) mixture of dithiothreitol:dithioerythritol.

matrixes produce different molecular ion adducts needed for confirmation of proposed molecular weights.

An alternative method for applying the high-acceleration voltage to the multisample probe tip was evaluated utilizing the VG coaxial FAB probe with a tip in the form of wedged spring. The coaxial probe is inserted until contact is made with the multisample probe tip, at which point the multisample probe is translated to sequentially analyze each sample. Further development of this approach was not pursued because it requires the simultaneous use of two high-vacuum inlets and careful positioning of the axial probe tip without bending the multisample probe tip off its perpendicular axis.

The mechanical engineering diagrams for the multisample FAB probe are available on request from the corresponding author.

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