Implementation of the Chemical Reaction Interface Mass Spectrometry Technique on a Hewlett–Packard Mass-Selective Detector

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A microwave-powered chemical reaction interface has been installed in a Hewlett–Packard gas chromatograph–mass spectrometer (GC-MS) system (5890 II gas chromatograph–5971 mass-selective detector). The technical details and optimization strategies are discussed. The evaluation of this new setup is presented, showing detection limits of 1 ng of ^{13}C-, ^{15}N-, and Cl-containing compounds with signal-to-noise ratios greater than or equal to 3. Selective detection was evaluated with a urine sample from a dog dosed with $^{15}N_3$-midazolam that had been previously analyzed by using a differentially pumped research-level quadrupole mass spectrometer. The results show that the detection of ^{15}N and Cl remains highly selective and the mass-selective detector gives comparable sensitivity to the larger instrument when the latter is operating over a conventional mass range. The capability for chemical reaction interface mass spectrometry can be easily accomplished with an inexpensive GC-MS system. *(J Am Soc Mass Spectrom 1994, 5, 765–771)*

Chemical reaction interface mass spectrometry (CRIMS) was introduced in 1982 [1] and since then has been demonstrated to be a useful analytical tool in drug metabolism, biological, and environmental applications. CRIMS combines selective detection of elements and their isotopes and conventional mass spectrometry in a single system involving few modifications to the instrumentation. Selective detection of ^2H, ^{13}C, ^{14}C, ^{15}N, S, and Cl has been reported [2–12]. The range of analytes includes drugs and drug metabolites, environmentally important substances, and endogenous compounds. Isotopic enrichments as low as 0.0044 atom % excess for ^{15}N can be measured [10]. Detection limits below 100 pg for sulfur- and chlorine-containing compounds have been achieved [7, 9].

The concept of CRIMS is to completely atomize an analyte in a microwave-induced helium plasma and then permit the atoms to recombine with a reactant gas. This produces an array of small gaseous species that are detected by the mass spectrometer (MS). The amounts of these newly formed species relate to the amount of the analyte and the number of each element that was present in the original structure. Any isotopic enrichment of the newly formed species reflects the isotopic enrichment of the analyte.

A schematic reaction sequence for CRIMS with SO_2 as the reactant gas (without fully considering stoichiometry) is

$$C_pH_qO_rN_sCl_t \rightarrow pC + qH + rO + sN + tCl \quad \text{(atomization)}$$

$$SO_2 \rightarrow$$

$$S + O \rightarrow O_2 + O_3 + SO + SO_3 \quad \text{(reactive species)}$$

$$C \rightarrow CO_2 + CO \quad \text{(CRIMS products of carbon)}$$

$$H \rightarrow H_2O \quad \text{(CRIMS product of hydrogen)}$$

$$O \rightarrow SO_2 + O_2 \quad \text{(CRIMS products of oxygen)}$$

$$N \rightarrow NO + N_2 \quad \text{(CRIMS products of nitrogen)}$$

$$Cl \rightarrow HCl \quad \text{(CRIMS product of chlorine)}$$

This set of products defines the analytical abilities of CRIMS with SO_2 as a reactant gas.

The useful species are CO_2, NO, and HCl. These three products show sensitive, selective, and linear detection characteristics and provide good isotopic information [2, 3, 5, 7–12]. Water is not a good analytical species because of adsorption, reactivity, and exchange. CO and N_2 are observed at the same nominal

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mass and are each of lower intensity than are CO_2 and NO, respectively. The oxygen contained in an analyte is presumably in equilibrium with the oxygen contained in the SO_2 reactant gas. Because the reactant gas is in substantial excess, neither the oxygen nor sulfur content of an analyte can be observed by using this particular reactant gas.

The proper operation of CRIMS requires that the three chemical species involved—the helium carrier gas, the reactant gas, and the analyte—have a correct relationship. The helium must be in considerable excess, because the energy available for the atomization reactions depends on the high ionization potential of helium. The reactant gas must be in low enough proportion that the helium plasma is not quenched. This is accomplished by viewing through a small hole that is incorporated in the microwave cavity the color and brightness of the ceramic tube in which the plasma is sustained. When the reactant gas is introduced at very low concentrations, the color changes from a yellow–orange color that is characteristic of helium to a whitish blue. As the SO_2 is increased, the color remains fairly constant, but the brightness dims. This dimming indicates quenching of the plasma, although some dimming is acceptable.

The capacity of CRIMS for analytes is determined by the level of reactant gas. To maintain the constant array of reaction products that are listed above, the reactant gas must remain in significant excess compared to the analyte. Whether the criterion is fulfilled can be assessed in several ways. The most direct method is to monitor an ion that comes from the SO_2 itself. Because there is a substantial amount of SO_2 present, the parent ion, m/z 64, is always off scale. Therefore, an isotope peak of smaller abundance, such as m/z 66, can be used. When the plasma is first ignited, the amount of reactant gas decreases somewhat because of the presence of stable decomposition products, such as SO, SO_3, O_2, and O_3, but remains stable thereafter except when large amounts of an analyte are eluting. Under these conditions, the SO_2 signal drops, which indicates that a percentage of the reactant gas is being used up by the analyte. We have found a better way to define good CRIMS conditions than to try to establish the acceptable level of this drop. Carbon disulfide, which is formed under reducing conditions in CRIMS, appears only when inappropriately large amounts of analytes enter the reaction chamber. The presence of this reduction product implies that the oxidation capacity of the reaction interface has been exceeded and that the analytical result is potentially flawed [2].

The most important attribute of CRIMS is that it provides a type of selective compound-independent detection that is usually achieved only with radioisotopes. The most obvious advantage of CRIMS is that radioisotopes are not needed. Another advantage is that CRIMS enables higher chromatographic resolution than most radioisotope detectors, where significant volume contributes to notable band-spreading. Finally, CRIMS has been shown to be more sensitive than in-line counting methods for ^{14}C [4].

The Hewlett–Packard mass-spectrum detector (MSD) is a highly popular gas chromatograph–mass spectrometer (GC-MS) system because of its small size, high performance, and low cost. Furthermore, the Hewlett–Packard 5890 GC is used in many other GC-MS combinations. However, CRIMS has not been adapted to a Hewlett–Packard system before. The development of a GC-CRIMS system based on the Hewlett–Packard GC-MSD should enable easier access to this simple yet powerful technique. The objective of this work is to implement CRIMS on a Hewlett–Packard MSD system, explain the optimization strategies, and evaluate its performance in comparison to other GC-CRIMS systems operating on more complex instruments.

Experimental

Apparatus

The mainframe of the instrumentation is a Hewlett–Packard (Palo Alto, CA) 5890 II GC–5971 MSD system. A chemical reaction interface (CRI) is installed in the GC oven (Figure 1), which consists of a microwave cavity, a 5-in.-long 1/4-in.-o.d., 3/32-in.-i.d. alumina tube (99.8% purity; Omega Engineering, Stamford, CT), and a 100-W 2450-MHz Model MPG-4 microwave power supply (Opthos, Rockville, MD). The alumina tube must be specified to 0.250 ± 0.002 in. to meet the tolerance of the Swagelok (Swagelok Corp., Solon, OH) fittings. The mass spectrometer itself was not modified.

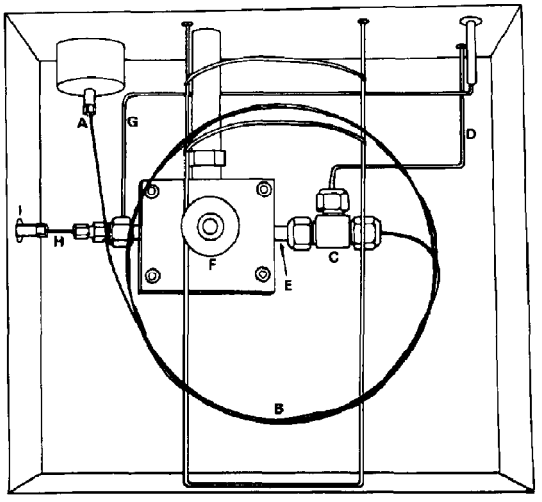

Figure 1. Front view schematic of the Hewlett–Packard 5890II GC oven with a CRI. A: GC injector port; B: GC column; C: Swagelok T; D: reactant gas line; E: alumina tube; F: CRI cavity; G: ignition wire; H: transfer line; I: MSD inlet.

Microwave Cavity. A newly designed stainless steel microwave cavity (Figure 2) was built by Vestec (Houston, TX). The major differences between the new design and the old cavities are that knife-edge seals were used to connect the endplates to the body of the cavity and the adjustable inner focusing cylinders are joined to the endplates with welded bellows rather than the previous design in which all of these connections were threaded. The knife edges and bellows provide a consistent electrical conduction path that solves a previous problem of unstable microwave power transmission to the plasma in the reaction chamber. Because the reaction chamber is continuously subjected to the extremes of heat and temperature cycling in GC-CRIMS, thermal stress caused warping and metal fatigue in the threaded design. This more complex design is not necessary with high-performance liquid chromatography (HPLC) CRIMS [11], where the cavity remains at room temperature.

Gas Chromatograph Oven Modification. To install the CRI cavity in the Hewlett-Packard 5980 II GC, a hole in the oven is needed to allow the cavity arm that brings in the microwave power to meet with the microwave power supply cable. One of the existing holes in the roof of the oven was widened by 2 mm, and the insulating glass fiber was removed. A 1/4-in.-thick 4- × 6-in. aluminum plate was mounted onto the top of the oven by using two existing prethreaded holes. The plate has a 1-in.-i.d. hole located in the center, on the top of which is mounted a 1/2-in. aluminum clamp with a 1/2-in.-i.d. aperture to hold the arm and fix the cavity in the oven.

A stainless steel Swagelok T (1/4 in.) is used to couple the capillary column, the alumina reaction tube, and the reactant gas line. Graphite/Vespel ferrules are used throughout, including no-hole 1/4-in. ferrules that are drilled out to accept the capillary column and the 1/16-in. line carrying the reactant gas. Uncoated deactivated fused silica tubing was used as the transfer line between the reaction chamber and the MSD ion source. The 0.32- and 0.53-mm-i.d. material was purchased from J & W Scientific (Folsom, CA) and the 0.35- and 0.40-mm-i.d. material came from Polymicro Technologies (Phoenix, AZ). The choice of the transfer line dimensions is detailed in the Results and Discussion section.

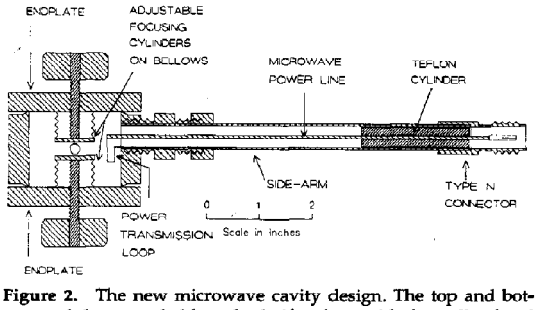

Figure 2. The new microwave cavity design. The top and bottom endplates are held to the knife edges with four allen-head screws (not shown). The bellows are welded to the inner surface of the endplates. The side arm is attached with a 1/2-in. to 1/2-in. Swagelok male connector. A Teflon cylinder with a 1/16-in. hole is used to keep the microwave power line centered in the side arm. This line is the only gold-plated component in the cavity assembly.

Sometimes the helium plasma ignites spontaneously and sometimes a high frequency generator (BD-40E, Electro-Technic Products, Chicago, IL) was needed to initiate ignition. A piece of bare steel wire was used to transmit the high voltage. One end of the wire gently touches the union joining the alumina tube and the transfer line, and the other end goes out through a ceramic insulator traversing the oven wall to avoid contact with metal (see Figure 1). The high frequency generator is adjusted to the lowest setting to make a spark and briefly touched to the ignitor wire to initiate the plasma.

The high voltage produced from these generators is accompanied by considerable stray rf energy that may affect computers and GC control boards that are part of the GC-CRIMS system. The video board seems especially susceptible. The screen image will disappear, requiring a reboot. Occasionally, another spark will reactivate the video board. The use of this particular high frequency generator markedly reduced the number of occurrences of digital system interruptions compared to the previously used Tesla coil device.

MSD Operating System. The Hewlett-Packard 5971 MSD mass spectrometer is operated by a Teknivent (Maryland Heights, MO) Vector 2 data acquisition interface and software using a 486-chip-based computer. The primary reason for use of this equipment rather than the standard Hewlett-Packard data system is because Teknivent provides the CALC function needed to generate enrichment-only chromatograms [2]. Two different selected-ion recording procedures were used. Most experiments monitored five masses: m/z 30, 31, 44, 45, and 76, with m/z 31 and 45 integrated 10 times longer than the other channels. The total cycle time was 1 s. For the combined ^{15}N- and Cl-selective detection experiments, m/z 30, 31, 36, and 44 were monitored with m/z 30, 31, and 36 integrated 2.5 times longer than m/z 44 using an 800-ms total cycle time.

Mass-Selective Detector Tuning and Calibration

It is necessary to manually tune and calibrate quadrupole mass spectrometers for CRIMS work. This is important, because the reactant gas produces huge ions at m/z 32 and 48 that interfere with ions used for monitoring nitrogen and carbon; that is, m/z 30, 31, 44, and 45. These ions must be completely resolved from interfering masses to measure correct isotope ratios and low isotopic enrichments. The tuning and resolution adjustments of the mass spectrometer should

be performed when the CRI is turned on, because the intensities of the target masses increase greatly. The calibration windows should be narrow (4 to 6 u wide) and the operator must expand the vertical scale to visualize the smallest peaks in the mass range so as to observe any overlap between nearby masses. The instrument will appear to be overresolved for major peaks when good baseline separations between large and small masses are achieved. Although the Hewlett–Packard MSD instruction manual recommends negative polarity, we experimented and found that, for us, changing the quadrupole polarity to positive dc improves mass resolution significantly. The scanning mode of data acquisition is calibrated normally with perfluorotributylamine, but selected-ion monitoring calibration is done without this standard. We carefully select the centroids of m/z 18, 28, 32, 40, and 44 as the calibration set.

Chemicals

The GC carrier gas is ultrapure helium from Air Products and Chemicals (Allentown, PA). A stainless steel diaphragm high purity regulator is used and the entire GC system, from the He tank through the MS, was preevacuated with a rough pump via a valved port on the He regulator prior to initially opening the He tank valve. This is necessary because air entrapped in gauges and controllers represents "virtual leaks" that may contaminate the tank or contribute air to the background and persist for extended periods.

Sulfur dioxide (Matheson, East Rutherford, NJ) was used as the reactant gas. The SO_2 flow is adjusted by a Granville-Phillips (Boulder, CO) series 203 variable leak valve. Because trace amounts of air or other gases affect the chemical reaction interface, the entire gas train from the tank to the variable leak valve was evacuated before use by rough pumping a valved port on the SO_2 regulator prior to initially opening the SO_2 tank. The SO_2 cylinder was specially processed by the manufacturer by first filling with the usual 4 lb of liquid SO_2 and then bleeding off half of the tank to purge the entrapped air.

Diazepam and $^{15}N_3$-midazolam were provided by Hoffmann LaRoche Inc. (Nutley, NJ). [2-$^{13}C_1$,1,3-$^{15}N_2$]-Caffeine was obtained from Cambridge Isotopes (Woburn, MA).

Sample Preparation

A previously described method was used to prepare the urine sample [12]. A 10-mL aliquot from the 0 to 48-h urine of a male beagle dog dosed orally with 5-mg/kg $^{15}N_3$-midazolam was filtered and incubated at pH 4.5–5.0 and 37 °C for 24 h with glucuronidase/sulfatase (Sigma, St. Louis, MO, type H1). The reaction mixture was subjected to solid phase extraction with a C18 Alltech (Deerfield, IL) Maxi-Clean cartridge, washed with 10 mL of water, and then eluted with a series of 2.0 mL of methanol solutions (25, 50, 75, and 100% in water), each collected separately. The eluates were dried with a stream of nitrogen and then derivatized with 50 μL of N-methyl,N-trimethylsilyl-trifluoroacetamide and 50 μL of acetonitrile at 100 °C for 30 min. As opposed to the prior chromatogram shown for this sample that included all four eluate fractions [12], this work examined just the 50% methanol eluate.

One microliter of sample was injected in the splitless mode and separated on a 30-M 0.25-mm-i.d. 0.10-μm film DB-5 capillary column (J & W Scientific). The column temperature was initially held at 80 °C for 1.5 min, programmed at 30 °C/min to 110 °C, then at 6 °C/min from 110 to 270 °C, and held for 10 min. The end of the column passes through the Swagelok T and is placed just inside the alumina tube. If the fused silica column is inserted into the plasma region, the coating will be degraded, which markedly elevates the baseline.

Results and Discussion

Optimization of Operating Conditions

The CRIMS chemistry and the general characteristics of optimization have been described in previous publications [2, 7, 9]. In this article, we emphasize those technically important details that have not been fully described.

Transfer Line Selection. The gas flow resistance of the transfer line and the helium flow rate determine the pressure inside the alumina tube in which the microwave-induced helium plasma exists. If the resistance is too low, the system will be at suboptimal pressure. If the density of reactant gas is too low and an insufficient number of collisions occur, the chemical reactions might not reach completion. The resulting array of products will not be analytically complete. On the other hand, if the resistance is too great, high pressure in the alumina tube will quench the plasma so that no chemical reactions occur. Furthermore, as the pressure increases, the interface increasingly becomes dead volume and the eluting peaks are broadened. With the usual 1-mL/min helium flow, our experience demonstrated that 15 in. of 0.53-mm fused silica tubing was consistently successful [2, 9]. A series of transfer lines with different dimensions (0.53, 0.40, 0.35, and 0.32 mm i.d.) were tested because the Hewlett–Packard GC-MS interface uses only 10 in. of transfer line.

Previous investigations [9] in this laboratory showed that m/z 76 (CS_2) only appears when the oxidizing capacity of SO_2 is exceeded by the amount of the analyte. The ratio of the signals at m/z 76 to m/z 44 (CO_2) can be used to monitor the chemistry in the chemical reaction interface. When the ratio of m/z 76 to m/z 44 is greater than 0.001, the result is discarded

because of the possibility of poor chemistry. The experiments show that when a 10-in. × 0.53-mm transfer line was used, the ratio of m/z 76 to m/z 44 was 0.009, whereas all other transfer lines provided a ratio less than 0.001. However, the peaks with the 0.35- and 0.32-mm transfer lines were broadened compared to the 0.40-mm tubing; thus the 0.40-mm transfer line was considered best for this system.

Alumina Tube. Another series of experiments was conducted by changing the inner diameter of the alumina reaction tube. Two sizes, 1/16 and 3/32 in. i.d., were tested. The data from the 3/32-in.-i.d. tube showed better ratios of m/z 76 to m/z 44 than the data from the 1/16-in.-i.d. tube at comparable experimental conditions, although the improvements did not exceed a factor of 2. Most of our other GC-CRIMS experiments used the 1/16-in.-i.d. tube, whereas the wider aperture of the 3/32-in.-i.d. tube is used to better capture the particle beam for HPLC-CRIMS. The 3/32-in.-i.d. alumina tube was used for the experiments that are described here.

Reactant Gas and Helium Flow. In addition to the transfer line resistance, there are several other factors that affect the chemistry in the reaction chamber. Two of the most important are the helium and the SO_2 flow rates. Figure 3 is an example that shows how these two factors influence the chemistry. Because the absolute flow of SO_2 was difficult to measure, the highly nonlinear scale reading from the Granville-Phillips variable leak valve that controlled the SO_2 flow was noted. Similarly, the column head pressure was used to represent the helium flow. From Figure 3, one can see that as the amount of SO_2 increases or the helium flow decreases, the ratio of CS_2 to CO_2 is decreased, which means that the oxidation chemistry in the chamber is more appropriate. As mentioned in the introduction, too much SO_2 in the reaction chamber quenches the plasma, as well as decreases the ion source efficiency of any mass spectrometer. Low He flow presumably increases the residence time of analytes in the plasma, thus facilitating complete reaction.

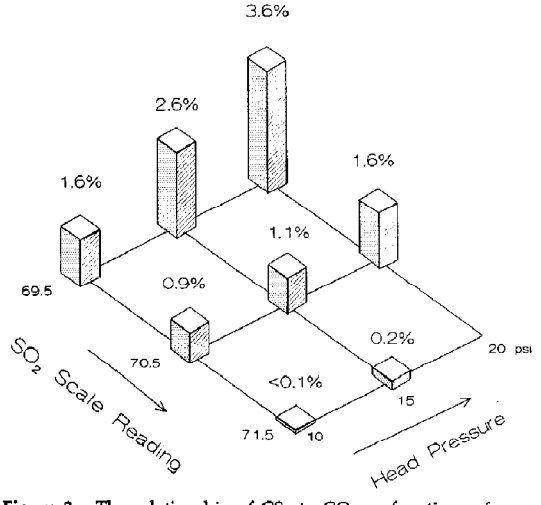

Figure 3. The relationship of CS_2 to CO_2 as functions of reactant gas (SO_2) and He flow rates. The magnitudes of these flows are not known, but are monitored, indirectly, for SO_2 by the scale reading on the Granville-Phillips variable leak valve and for He by the regulator pressure.

The flow resistance of the analytical capillary column is also important for the best CRIMS performance. Because the output end of the column is at vacuum, the head pressure can fall below 1 atm if the resistance of the column is too low. When this happens, backflow from sources of air, such as septum punctures, splitters, and purges, increases the amount of background. We have found that a 0.25-mm-i.d. column of at least 30 m is needed. Such a column requires greater than 14-psi head pressure to generate a 1-mL/min flow with atmospheric output pressure, which means that more than a 1-atm pressure drop will exist when the outlet is at vacuum. Wider bore and/or shorter columns may not operate properly.

The tubing that introduces reactant gas from the variable leak valve to the reaction chamber is another variable in the instrument setup. Two sections of stainless steel 1/16-in. capillary tubing (each 20 in. long; one 0.01 in. i.d. and the other 0.03 in. i.d.) were tested. The smaller diameter tubing required a longer time to reach equilibrium flow after adjusting the valve than the wider bore capillary. The peak widths were identical, which indicates that the wider tubing does not introduce significant dead volume into the system. We recommend wider inner diameter tubing for the introduction of reactant gas to the reaction chamber.

Reactant Gas Purity. Liquified gases such as SO_2 can have substantial head space air contamination. In this application we used the gas phase, which contains a disproportionate amount of impurity. Our first improvement was to have the tanks prepurged with helium, which reduced O_2 and N_2 background severalfold. However, low levels of nitrogen-containing analytes were still masked by a high baseline. The current procedure of bleeding off half the tank represents a further improvement. With the same experimental conditions and a half-bled tank of SO_2, the background of m/z 30 (NO; a CRIMS product of nitrogen impurity) was reduced fourfold compared to the He-purged SO_2 tank.

Selective Detection of ^{13}C, ^{15}N, and Cl

To show the ability of selective detection of isotopes and elements by CRIMS, caffeine labeled with one ^{13}C and two ^{15}N, and diazepam were chosen as the model compounds. With 1 ng of labeled caffeine, the signal-to-noise ratio for ^{13}C (detected as enriched $^{13}CO_2$) and

^{15}N (detected as enriched ^{15}NO) were 3 to 1 and 5 to 1, respectively. The observed $^{13}CO_2$ and $^{12}CO_2$ were measured at m/z 45 and 44, and the observed ^{15}NO and ^{14}NO were measured at m/z 31 and 30, respectively. The enriched $^{13}CO_2$ and ^{15}NO were calculated using the following equations with the CALC function of the Teknivent Vector 2 data system:

$$\text{enriched } ^{13}CO_2 = \text{observed } ^{13}CO_2 - \text{observed } ^{12}CO_2 * 0.0119$$

$$\text{enriched } ^{15}NO = \text{observed } ^{15}NO - \text{observed } ^{14}NO * 0.00403$$

The CALC function allows a user-generated algebraic manipulation of one or more masses to generate a derived chromatogram that can be acted on by the routine chromatographic functions of the data system. In the calculation, the natural abundances of ^{13}C, ^{17}O, and ^{15}N were all considered, and the theoretical abundances listed above correctly produced a blank CALC chromatogram following unenriched caffeine injections. This indicates that ^{14}NO and ^{15}NO are the only species produced by the analyte at m/z 30 and 31. With 1.0-ng injections of diazepam, the signal-to-noise ratio with m/z 36 ($H^{35}Cl$) as the detection channel was 3 to 1.

Application to a Drug Metabolism Study

To compare the ability of CRIMS on the Hewlett–Packard MSD with CRIMS on the Extrel C50/400 (Extrel Corp., Pittsburgh, PA)—a larger, differentially pumped research-quality quadrupole MS—the selective detection of Cl and ^{15}N was performed on a urine sample from a dog dosed with ^{15}N labeled midazolam (a sample that has already been the subject of a full investigation [12]). Figure 4 shows three chromatograms of the dog urine extract. The upper chromatogram is m/z 44 ($^{12}CO_2$), showing the general detection of organic compounds. The lower two chromatograms show selective label detection. As was previously demonstrated [12], despite the complexity of the urine sample, the ^{15}N and Cl channels were highly selective and matched each other. The most abundant labeled peak was identified as α-OH midazolam, in an amount we compute to be 60 ng. The two smaller peaks, each of which contain less than 10 ng of material, that elute earlier are newly discovered metabolites of midazolam [12].

We tried to be careful in our comparison of the two different CRIMS setups. The performance of our Extrel quadrupole depends significantly on what radiofrequency power supply is used. The Extrel instrument was purchased with a special 2.1-MHz rf power supply, which gives an upper mass of 200 u, that maximizes low mass transmission and resolution. It also has the conventional 1.2-MHz 800-u mass range rf

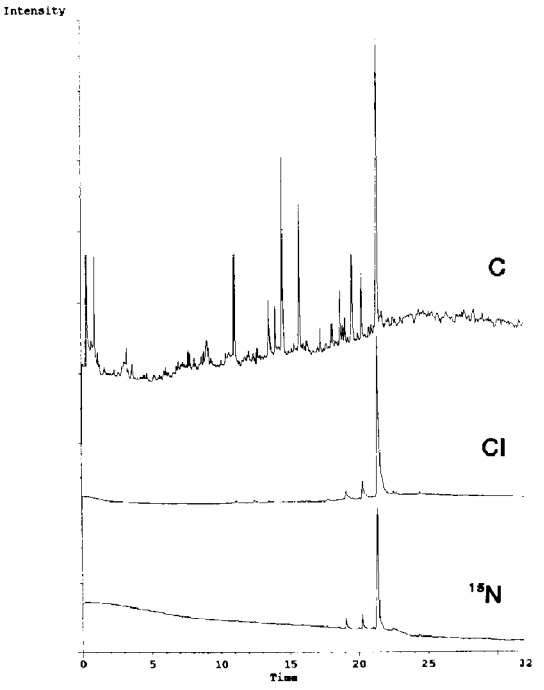

Figure 4. CRIMS chromatograms of a dog urine sample. The top trace represents total carbon content, the middle trace monitors chlorine, and the lower trace shows enriched ^{15}N. The Cl and ^{15}N traces are magnified 10 and 2 times, respectively, compared to the carbon trace.

power supply. With the 2.1-MHz configuration, the detection limit of m/z 36 (HCl) was 50 pg of diazepam ($S/N \geq 3$). This is a much better result than the Hewlett–Packard MSD, where the detection limit was 1 ng of diazepam with comparable conditions for data acquisition. The Hewlett–Packard MSD has a mass range of 0 to 650, and the low mass region is further discriminated against because of the ion source magnet that collimates the electron beam. When the Extrel was tested with the 800-u supply, the result for chlorine showed that the Hewlett–Packard MSD provided a threefold better signal-to-noise ratio than did the Extrel MS.

Routine Use

Following evaluation as described above, the GC-CRIMS system was used to conduct investigations of new CRIMS reactant gases. As such, we have acquired few data regarding precision with this instrument. With the Extrel instrument the precision of CRIMS for isotopic abundance can be as low as 0.6% relative standard deviation (RSD) [10], and we measured the C to Cl ratio for three benzodiazepines with RSDs of 2 to 7% [9]. Our current work involves developing phosphorus-selective detection with the Hewlett–Packard instrument. The average error for replicate measure-

ments in a standard curve of two phosphorus-containing compounds was less than 5% over most of the concentration range and the correlation coefficient, r^2, was 0.997. Although these numbers are not exactly comparable to the RSDs cited above, they indicate to us that the analytical performance of the Hewlett–Packard MSD-CRIMS system is consistent with good quantitative analytical work.

Conclusions

A GC-CRIMS system has been created with a Hewlett–Packard 5890 II GC–5971 MSD. Few modifications to the GC and no modifications to the MSD were required. The evaluation shows that the new setup provides good sensitivity for the selective detection of isotopes and elements, in this case, ^{13}C, ^{15}N, and Cl. The attempt to apply the new GC-CRIMS to a real drug metabolism study with a urine sample from a dog dosed with $^{15}N_3$-midazolam was successful, and again proves that CRIMS is a powerful analytical technique that now can be carried out with a simple GC-MS system.

Acknowledgments

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