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In the Classroom

Fourier Transform Infrared Spectroscopy Across the Chemistry Curriculum

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In 1994, the Chemistry department at Huntingdon College was awarded a National Science Foundation Instrument and Laboratory Improvement grant for the acquisition of an FTIR spectrometer. The instrument has supported improvements in lecture and laboratory courses throughout the chemistry curriculum and promoted a “Jan-Term” minicourse entitled “FTIR Spectroscopic Analysis.” Undergraduate independent study research projects now emphasize laboratory work, students are making formal presentations of their work at scientific meetings, and publications including student authors are in preparation.

It has become essential, therefore, that undergraduate science majors gain experience in spectroscopic analysis using modern instrumentation.

Introduction

Science laboratories involved in research, industry, environmental analysis, and other scientific endeavors commonly use modern, computer-interfaced instrumentation to analyze minute samples quickly and accurately. It has become essential, therefore, that undergraduate science majors gain experience in spectroscopic analysis using modern instrumentation. The NSF Division of Undergraduate Education recognized these influences and requirements in their report entitled “Innovation and Change in the Chemistry Curriculum” [1].

Although Huntingdon is a small college (about 700 students), the chemistry department graduates an average of 5 to 12 majors every year. The support of the National Science Foundation and the College has added considerable impetus to improvements in an already strong chemistry program. The recent acquisition of an FTIR spectrometer is providing students with valuable laboratory experiences in modern spectroscopic analysis and has increased the impact of other changes which promote spectroscopic analysis throughout the chemistry curriculum.

Improvements in Freshman Lecture and Laboratory Courses

In order to provide greater emphasis on spectroscopic analysis throughout the chemistry program, basic principles of spectroscopy are now introduced in the General Chemistry lecture sequence. The topic fits well because of its historical success in explaining atomic line spectra and the subsequent development of the theories of atomic and molecular structure. Absorption and emission spectroscopy are discussed and compared as well as the types of atomic and molecular excitations that are promoted by the various electromagnetic energies used for irradiation. This pedagogical approach provides a more tangible avenue to the development of the theory of electronic structure of atoms and the concepts of orbitals and electron configurations than the more traditional, but abstract, freshman-level quantum mechanics. A paper which addresses this approach in detail has been published recently [2].

Course components in spectroscopy have been added to the General Chemistry laboratory sequence. Students in the General Chemistry laboratory now experience spectroscopy firsthand by analyzing atomic line spectra (e.g., of hydrogen, sodium, etc.) with spectroscopes. A brief discussion of practical applications, such as the analysis of gases on distant planets and the observation of Doppler shifts due to the expansion of the universe, can be used to introduce the laboratory exercise. Students progress to using Spectronic-type colorimeters for environmentally pertinent analyses such as vitamin-B mixtures and food colorings. These provide interesting and relevant instruction in calibration, the Beer–Lambert law, and quantitative analysis.

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FTIR spectroscopy is demonstrated in two separate experiments, one in which students identify unknown organic liquids by their boiling points, and another in which solid unknowns are identified by their melting points. In each case students verify their initial conclusions by observing as the instructor performs an FTIR analysis of their unknowns, followed by a spectral library search-and-match while describing in general terms what is being done. This sets the stage for hands-on use, which begins after Freshman-level courses.

Improvements in the Organic Lecture and Laboratory Courses

Spectroscopy is now taught in the first semester of the Organic Chemistry lecture sequence from about the fifth week onwards. This pedagogy reinforces the study of molecular structure, functional groups, isomerism, and inductive and mesomeric effects on the strength of organic acids and bases. By bringing organic spectroscopy into the first semester [3, 4], student expertise in analyzing spectroscopic data to solve problems has improved significantly, because they are continuously exposed to spectroscopy throughout the year as they study the properties of each new functionality in turn.

One advantage for students attending small colleges is the personal level of instruction that they receive in small classes that are taught by professors, rather than graduate students. Additionally, in smaller laboratory courses students are able to use the instruments more often and thereby gain considerable hands-on experience in sample preparation, data manipulation, library searching and matching, and saving and printing spectra. After the Freshman year, most chemistry courses at Huntingdon have 5 to 14 students; therefore, there are few (if any) time constraints in restricting consistent use of instruments by the students in their experimental methodology. This is a major advantage offered by modern FT instruments, for which analysis times are a fraction of those required for the older (dispersive) instruments. Although time must be set aside to train students to use computer-interfaced instruments, most students are already familiar with using computers, Microsoft® Windows™, etc., and it generally takes only about one laboratory session for students to learn to run an FTIR spectrum.

The content for the introductory session on FTIR spectroscopy in the Organic laboratory is supplied to students in outline form, as shown in Figure 1.

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Introduction To FTIR Spectroscopy

Organic Chemistry I Laboratory

1. Review of information contained in an IR spectrum
    a. functional group region
    b. fingerprint region
    c. location (in wavenumbers) of bands

2. Overview of instrument hardware
    a. helium/neon laser
    b. IR source
    c. interferometer
    d. sample compartment
    e. detector

3. Sample preparation
    a. need for salt plates and the types that are available
    b. how to use and clean salt plates
    c. liquid-film technique for nonvolatile liquids

4. Running the analysis
    a. basic software commands
    b. number of scans and spectral resolution
    c. background and sample collection
    d. baseline correction and smoothing
    e. labeling important bands
    f. library searching and matching
    g. printing

FIGURE 1. OUTLINE FOR THE INTRODUCTORY SESSION ON FTIR SPECTROSCOPY SUPPLIED TO STUDENTS IN THE ORGANIC CHEMISTRY LABORATORY.

Part 1 is a review of material that has already been covered in greater detail in the lecture course. For part 2, the instrument casing is opened to reveal the various components in the interior. Although instrument hardware is treated in general terms in the Organic courses, it is taught in greater detail in the Instrumental Analysis course (see below); thus, the helium-neon laser is located and its function described as a multiple

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internal standard that allows addition of multiple scans, producing increased signal intensity and decreased random noise (Connes' advantage) in the resulting spectrum.

Parts 3 and 4 are the practical aspects of FTIR analysis that are not part of the lecture course and must be discussed in some detail in this laboratory session. Students are shown different types of salt plates (potassium bromide, sodium chloride, zinc selenide, etc.) and a spectrum (framed wall poster) illustrating where the different materials absorb infrared radiation. The properties of the various salt plates are described with particular emphasis on their sensitivity to water.

In the first session the simplest method of analysis, the preparation of liquid films on sodium chloride or potassium bromide plates, is described. The instructor runs an analysis of a suitable nonvolatile liquid such as benzaldehyde, demonstrates how to set the spectral resolution (8 cm^{-1} is quite sufficient for liquids), the number of scans, run the background scan, and run the sample scan. The default settings are entered ahead of time so that students will not need to make adjustments until they have gained more experience with the instrument. Students are taught how the computer collects data and the meaning of the spectral resolution setting. This is an important topic that is related to the number of data points and the overall quality of spectra. The spectral resolution also affects any comparisons made with computer spectral libraries (see below) where spectra generally have a resolution of 8 to 16 cm^{-1} and, therefore, may be lacking in the detail provided by the sample spectrum. This may result in the omission of small peaks and shoulders in the library spectrum being compared, and the absorption band maxima may occur at slightly different wavenumber frequencies. Students also learn how FTIR augments the signal-to-noise ratio and decreases analysis time by the multiplex nature of the FTIR signal (Felgett's advantage) and the greater throughput reaching the detector (Jacquinot's advantage).

With the background spectrum on the monitor screen, absorption bands due to water and carbon dioxide are pointed out. The absorption bands in the resulting sample spectrum are assigned to structural features of benzaldehyde, and spectral manipulations for baseline correction and smoothing are demonstrated. These processes and the spectral artifacts that they introduce are easier for students to understand because they have already learned how the computer collects and stores data. Students are taught that baseline correction may arbitrarily delete real absorptions, and that smoothing introduces an artificial averaging of absorption intensities. They are cautioned, therefore, to do as

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little manipulation as possible and to avoid any manipulations at all when quantitative calculations are required. The baseline correction process (carried out on the absorption IR spectrum) leads to the relationship between transmission and absorption spectra and the linear relationship between concentration and absorbance (the Beer–Lambert law).

After applying software commands to label important bands with their absorption frequencies (in wavenumbers), students are shown how to set up and search the various libraries of stored spectra to obtain the best match. Once again, the type and method of computer data storage is described so that students will not rely too much on computer matching. Students are instructed to compare the actual spectra of computer hits with the sample being analyzed.

Finally, each student is given one or two drops of an unknown organic liquid for which, under supervision, they must run the IR analysis, label the major bands with their absorption frequencies, print a hard copy of the spectrum, and carry out a library search-and-match. They are also required to assign the observed bands to functional groups and note the five best computer hits.

After analyzing five or six samples in subsequent laboratory sessions, students gain sufficient expertise to run the instrument with minimal supervision. Log books are maintained to keep a record of any instrumentation problems that may occur, and students are encouraged to consult the professor for any help they require.

Students learn how to make potassium bromide pellets for solids analysis in subsequent experiments—FTIR analysis is now a component of most experiments in the Organic laboratory. Other sampling techniques, such as gas analysis, horizontal attenuated total reflectance (HATR), and solutions, are used mostly in other courses (see below).

Improvements in the Instrumental Analysis Course

The Instrumental Analysis course comprises lecture and laboratory components. The lecture component [5a] has been updated and expanded to include the following:

1. Background

IR region of the electromagnetic spectrum; vibrational excitation modes and resulting

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absorption; requirement that observable bonds have a dipole; the relationship between band intensity and strength of the dipole.

2. Dispersive Instruments
Internal components: source, beam splitter, monochromator dispersion of transmitted beam into sequential wavelengths focused on the detector.

3. FTIR Instruments
Internal components: IR source, Michelson interferometer, beam splitter, helium-neon laser source and detector, deuterated triglycine sulfate (DTGS) pyroelectric detector.

4. FTIR Advantages
Resolution enhancement: Fellgett's (multiplex) advantage; better signal-to-noise ratio: Jacquinot's (throughput) advantage, Connes' advantage (presence of multiple internal standards due to the helium-neon laser scanned spectrum).

5. The Interferogram
Centerburst and wings.

6. The Fourier Transform
Cooley-Tukey algorithm (fast Fourier transform).

7. Phase Correction
Correction of unsymmetrical centerburst due to filtering of detector noise and other electronic sources.

8. Resolution and Apodization
Length of mirror stroke, multiple scans and signal-to-noise ratio, weighting of the interferogram (triangular, Happ–Genzel, Norton–Beer apodization) to remove pods.

9. Zero Filling and Smoothing
Mathematical interpolations to improve band conformation and signal-to-noise ratio.

In the laboratory component of the Instrumental Analysis course, students are instructed in the use of a 10-cm gas cell, and then they carry out a spectral analysis of a fluorocarbon. This experiment and the laboratory component of this course is still being developed to reflect a major emphasis on environmental analysis and protocols. Trial

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experiments under consideration include: (a) analysis of opaque coatings by HATR [6], (b) calculation of molecular weight of polymers by quantitative analysis of the ratio of a repeating functionality versus the concentration of the sample (for a soluble polymer) or by determination of the ratio of an end group to a repeating functionality, (c) determination of the signal to noise ratio as a function of the number of scans, (d) determination of a mixture of hydrocarbons. Still, quantitative analysis by FTIR spectroscopy is not a simple matter [7], and such experiments may be better suited for advanced courses or individual study.

Students are taught that sampling procedures affect the resulting analytical data and, therefore, must be selected with care. For example, the morphology of a polymer cannot be investigated if the polymer is subjected to heat or pressure or if it is dissolved as part of the sampling procedure. The sample matrix also influences the analytical results. For example, the polarity of a solvent will affect the degree and type of hydrogen bonding in a sample solution.

This course generally has 5 to 14 students enrolled. This allows the instructor to assign a different experiment to each student. Additionally, students will be given problems in which they must work as a team. Both approaches are valuable training for future careers in science and offer the opportunity to integrate other techniques and methods. For example, in the analysis of a mixture of hydrocarbons, gas chromatography is used as a complementary analytical method, and the students use spreadsheets or databases to correlate their results, make graphs, etc.

Improvements in the Physical Chemistry Lecture and Laboratory Courses

In the Physical Chemistry courses components [8] have been added to illustrate the symmetry of simple gaseous molecules, the resulting rotational and vibrational excitation selection rules, and the structural information that can be deduced from the IR spectra (e.g., force constants of bonds and bond length).

FTIR Spectroscopic Analysis—a Two-Week MiniCourse

Two-week minicourses are offered at Huntingdon College during the interim period between the Fall and Spring semesters. In 1996, a “Jan-term” course entitled “FTIR Spectroscopic Analysis” was taught. Jan-term courses do not count towards the major, but they do carry academic credit. In general, there are few requirements for the course other than attendance and assignments. The courses are graded on a pass/no credit basis. These courses provide students with an opportunity to experience subject matter out of sheer curiosity; they offer faculty a chance to teach and stimulate students in areas of their own interest and explore new pedagogical methods.

The FTIR minicourse is designed so that any student with sufficient background and understanding of the basics of molecular structure is able to handle the material. The intention is to cover the fundamentals of IR spectroscopy using interesting, contemporary applications in a relaxed, informal setting, avoiding highly technical and mathematical approaches in favor of a broader, pictorial understanding of the subject matter [5b].

Eight students (this was the limit) attended the class, which met for three to four hours every day for two weeks (weekdays only); about one hour of each class was for theory and discussion and, after a short break, the remaining two hours (sometimes longer) focused on laboratory applications. In order to stimulate student interest in chemistry, classroom lectures were not rigidly controlled, but were open to discussion of the students' interests, questions, and opinions with regard to societal issues. The laboratory sessions emphasized environmentally-related analyses using the "discovery" approach. This approach provides those students having little or no background in IR spectroscopy with an exciting and motivating learning experience. Overall, the plan provided students with an opportunity to study and experience interesting applications of modern scientific instruments, gain a conceptual understanding of FTIR spectroscopy, and learn about some chemistry topics of environmental significance.

The structure and content of the daily meetings are detailed as follows.

Day 1

Classroom Session

This session discusses the properties of electromagnetic radiation: wavelength, velocity, frequency, wavenumbers; the basic theory of vibrational excitation by infrared radiation; appearance of the IR spectrum; and the Lewis structures of typical organic functional groups.

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Laboratory Session

The students are shown how to run a liquid-film IR analysis, the use and care of salt plates, properties of different salt plates, how to set the number of scans and the spectral resolution, baseline correction, the labeling of important absorption bands, and how to print the spectrum. They are given small liquid samples of compounds with similar carbon structure, but different functional groups, to analyze and assign the functional group vibrations by comparison; teamwork was encouraged. Examples of compounds are selected from the following sets: (a) octane, octanol, octanone; (b) toluene, benzyl alcohol, benzylamine, benzyl cyanide; (c) butanol, butylamine, butyric acid.

Day 2

Classroom Session

Students present the previous day’s analyses. Then, the absorption frequencies of common functional groups and the hardware arrangement and its comparison to dispersion IR spectrometers are presented by the instructor.

Laboratory Session

How the computer stores spectral data, spectral libraries, and searching and matching, spectral overlay for comparison, and the making of potassium bromide discs for solid sample analysis are discussed. Each student is given a solid or liquid sample of an unknown that they analyze, assign absorption bands to functional groups, and find the best spectral-library match by computerized searching and matching. The students are told that all the compounds are of environmental or biochemical significance. After identifying the compound by spectral searching and matching, they look up the structure of the compound, note the major uses of the compound, and discuss its environmental or biochemical significance. Students are referred to *The Merck Index* [9] as a major source for this information. Examples of compounds include nicotine, benzoic acid, phenol, caffeine, cholesterol, ascorbic acid, camphor, and styrene.

Day 3

Classroom Session

Students make presentations of the spectral characteristics and environmental aspects of the previous day’s laboratory work. The discussions following the presentations are open-ended and promote consideration of contemporary issues such as: nicotine addiction and the controversy surrounding allegations of added nicotine to tobacco products, the addition of stimulants (caffeine) to beverages, the role of cholesterol in

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heart disease and its biological significance in steroid biosynthesis, the benefits of vitamin C as an antioxidant and anticancer agent and Linus Pauling's postulates regarding resistance to the common cold, other contributions of Linus Pauling including the elucidation of the structure of proteins, the historical use of camphor as an antiseptic, and the industrial production of massive quantities of styrene and polystyrene in terms of the societal and environmental balance between improvement of the quality of life and harm to the environment. Remaining time is devoted to how FTIR instruments differ from dispersive instruments and the Michelson interferometer and how it works.

Laboratory Session

The Instrument casing is removed to reveal the moving mirror of the interferometer, the helium-neon laser is located, and the laser is tuned. Students see the appearance of the interferogram. Analysis of solutions and the selection of solvent is presented. The preceding topics prepare students for the next classroom meeting. Laboratory work consists of preparing a range of solutions with different concentrations of benzyl alcohol in carbon tetrachloride to examine qualitatively the relative ratio of free to hydrogen-bonded hydroxyl as a function of concentration [10].

Day 4

Classroom Session

The previous day's laboratory work is discussed. Hydrogen bonding and its importance in the structures of proteins and nucleic acids and environmentally significant uses of benzyl alcohol are presented. The discussion about how an FTIR spectrometer works, the appearance of an interferogram, and how an interferogram becomes a spectrum is continued.

Laboratory Session

Pharmaceuticals are analyzed. Student partner teams are given samples or pills of Advil®, aspirin, barbituric acid, and Tylenol®. They are shown how to carry out a simple extraction with an organic solvent, followed by filtration to remove inorganic substances, and evaporation of solvent. After IR analysis, students assign absorption bands, carry out a spectral search and match, then consult *The Merck Index* [9] for compound structures and physiological properties. Students define LD_{50} and note the corresponding values.

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Day 5

Classroom Session

Students present and discuss the previous day's results. An interesting and appropriate discussion of the physiological and societal impact of drug use and abuse usually ensues. The remaining time is devoted to a continuation of how an FTIR spectrometer works: sources, beamsplitters, and detectors.

Laboratory Session

Analysis of polymers: students are shown how to cast films and the heat-and-pressure method. Students carry out analyses of various polymers obtained from commercial goods (e.g., from 2-liter soda bottles) by pressing them using the potassium bromide press. The structure-property relationship of polymers is discussed to emphasize the destructive effect of these sampling methods on polymer crystallinity and morphology.

Day 6

Classroom Session

The previous day's results are discussed; comments are made on polymer uses and the societal and environmental impact of plastics. Spectral manipulations are presented: subtraction, baseline correction, smoothing, derivatives, and library searching. More complicated manipulations such as deconvolution and curve fitting are omitted.

Laboratory Session

Analysis by HATR is demonstrated using a sample of lipstick. Its spectrum is manipulated using baseline correction and smoothing. The first derivative spectrum is obtained and discussed. A full-spectrum library search reveals good matches with various fatty acid esters of glycerol. A library text search brings up a spectrum of "lipstick base" which is subtracted from the sample lipstick. Students carry out HATR analyses of other lipsticks and opaque materials.

Day 7

Classroom Session

Selection of sampling technique is discussed. Students were asked during a previous session to bring in a sample of any material of interest for FTIR analysis. The entire class is devoted to an open discussion of how best to analyze the materials that are brought in. Typical samples are: paint, carpet fibers, hair, nail polish, green leaves, ear wax, body lotion, and plastic grocery bags. These generate a lot of interest and some

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amusement. All the samples are considered by the students and questions lead to related topics in analytical chemistry such as forensic science, analysis of biological tissues, and environmental sampling.

The instructor does not participate in the eventual selection of the sampling method, except to answer questions and to make sure that the integrity of the salt plates is not compromised.

Laboratory Session

After choosing the most appropriate sampling method in the classroom, students analyze their samples, carry out spectral manipulations, and print the resulting spectra.

Day 8

Classroom Session

Students present their spectra and discuss the results. Because the instructor avoids helping the students run samples, except to answer questions, some analyses give very poor results. For example, an analysis of leaves was carried out by HATR because this was the only technique available in which the salt plates (zinc selenide) would not be destroyed by the water of the crushed leaf. The poor data resulted because intimate contact is required between the sample and the HATR cell window for a successful analysis, and the leaves were not sufficiently ground. Absorption due to water was an added complication because a spectral subtraction of water had not been performed, even though the students had recognized the need to use zinc selenide plates. Interestingly, analyses that do not turn out well prove to be the most instructive. In the case of the leaves, the possibility of presampling procedures, such as crushing the leaves and obtaining an extract, can be pointed out. The second half of the class is a continuation of sampling techniques: gas analysis, common gas cells, comparison of spectra of gases with those of liquids and solids, rotational transitions, and gaseous analysis by FTIR in environmental monitoring.

Laboratory Session

The class is assigned the problem of obtaining an FTIR spectrum of car exhaust. After collecting the sample in a 10 m gas cell, the analysis is run using high resolution (2 wavenumbers) and 64 scans. With guidance, students use the “zoom” software feature to maximize the rotation-vibration transitions due to carbon monoxide. Results are

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excellent. The overall mixture and the carbon monoxide expansion are printed for subsequent classroom analysis.

Day 9
Classroom Session
The carbon monoxide spectrum and the processes that give rise to P and R branches are discussed. The class continues discussing sampling methods including Diffuse Reflectance Infrared Fourier Transform Spectroscopy (DRIFTS).

Laboratory Session
Students run spectra of liquids or solids and compare them to their vapor-phase spectra. Samples of commercial, physiological, and environmental significance are selected. Students consult *The Merck Index* [9] for this information. Samples include toluene, benzene, methylene chloride, chloroform, ethyl acetate, *p*-dichlorobenzene, methyl *t*-butyl ether, and benzaldehyde.

Day 10
Classroom Session
Students present their results. This last class concludes with a summary of the course and the advantages and uses of FTIR spectroscopy as a powerful analytical tool.

The class was invited to comment on the course and they were all extremely positive.

Improvements in Undergraduate Research and Independent Study

The Council on Undergraduate Research affirms that research projects are unequaled for enhancing student abilities in critical thinking, design and execution of experiments, searching the literature, and developing oral and written communication skills. Such opportunities exist in departmental course offerings entitled “Independent Study in Chemistry” and “Honors in Chemistry.”

Before the acquisition of the FTIR spectrometer, all the chemistry projects were restricted either to library searches and the writing of a formal term paper or to independent study from a prescribed text with scheduled weekly meetings, homework, and tests. These projects rarely involved any laboratory work at all. Very few students (less than one per year) requested this option to partially fulfill the requirements for the chemistry major.

Since the FTIR spectrometer was obtained (less than two years ago) the high level of student interest in FTIR spectroscopy has been most evident from the number who have been requesting undergraduate projects and from ensuing developments. Highlights are listed below.

- Six students have carried out research projects with major emphasis on laboratory work and use of the spectrometer.

- Students now are presenting their work at scientific meetings: one student won the award for the best presentation by an undergraduate in the Science Education section at the Alabama Academy of Science meeting (University of Alabama-Birmingham, March, 1995); another student gave a poster presentation at the National ACS meeting in Orlando (August, 1996).

- A student voluntarily (without course credit) carried out a significant amount of work to contribute to this latter project presented in Orlando.

- Three publications that include student authors are in preparation.

- Publication of these projects will result in new experiments suitable for inclusion in undergraduate chemistry courses.

- Two more students are doing research projects in the present semester.

Summary

The acquisition of an FTIR spectrometer has promoted significant improvements throughout the undergraduate chemistry curriculum at Huntingdon College. The enhancement of student experiences in the practice of science using modern technology has had a profound, positive effect on their degree of expertise and their understanding of contemporary scientific and related societal issues. The instrument has become a focal point of the chemistry curriculum and has sparked student interest and initiative in carrying out research.

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