Use of Isomation in Ultimate Precision Spectrophotometry.

ARTICLE *in* ANALYTICAL CHEMISTRY · JULY 1965

Impact Factor: 5.64 · DOI: 10.1021/ac60227a041

2 AUTHORS, INCLUDING:

Chris Enke
University of New Mexico
198 PUBLICATIONS **4,646** CITATIONS

Use of Isomation in Ultimate Precision Spectrophotometry

Louis Ramaley[1] and C. G. Enke, Frick Chemical Laboratory, Princeton University, Princeton, N. J.

Ultimate precision spectrophotometry as described by Reilley and Crawford (*2*) involves the use of two standard solutions to set the 0% and 100% transmittance readings of the spectrophotometer. The full scale reading of the spectrophotometer is used for a much narrower concentration range than usual, and precision is thus increased. The method requires the two standard solutions mentioned above and several standard solutions with intermediate concentrations for the preparation of a calibration curve since the absorbance as read from the spectrophotometer is not a linear function of concentration. The 0% and 100% T standards have to be carefully chosen by extensive trial and error if maximum use is to be made of the capability of the instrument to expand the readout scale.

The method described below replaces the standard solutions and calibration curve with one standard solution and isomation. The instrument can be easily used to the maximum of its capabilities.

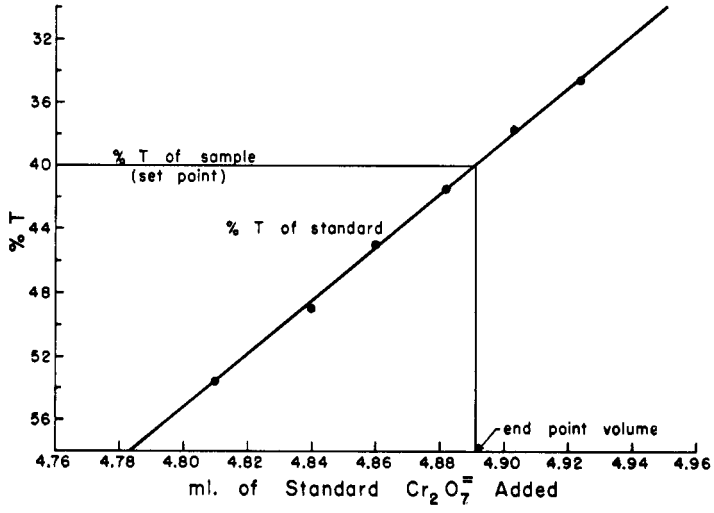

Figure 1. Isomation curve of sample containing $Cr_2O_7^{-2}$ vs ml. $Cr_2O_7^{-2}$ stock solution

Instrument adjusted to read 40% T with sample in light beam

METHOD

The sensitivity of the spectrophotometer is set to its highest value. The cell containing the sample is placed in the light beam of the instrument and the dark current control is adjusted until the spectrophotometer readout indicates 0% T or until it is no longer possible to adjust the dark current control. The slit width is then adjusted until the readout indicates about 90% T.

If there is no further adjustment possible for the dark current control, this is the final setting for the measurement. If some dark current adjustment remains, the control is adjusted until the readout is 0% T or no further change is possible. The slit width is again adjusted until the reading is about 90%. This procedure is continued until the dark current or slit width can no longer be adjusted.

The highest precision is obtained when the sample is set to read 100% T, but an end point in an isomation where the standard is increasing in concentration is passed over at this reading. With instruments which are capable of only a small scale expansion, the final reading or set point for the sample should be near 90% T to obtain the highest precision. However an instrument which has a large dark current offset and high scale expansion capability should be adjusted for a set point near midscale since the danger of overrunning the end point is greater and the relative gain in precision by operating near 90% T is less.

The spectrophotometer is now adjusted to give nearly the maximum precision of which it is capable for the particular sample in question. The 0% and 100% T readings correspond to definite concentrations, but these are unknown.

A cell containing an accurately known amount of the solvent is now introduced into the light beam. A standard solution of the sample substance is added to the cell until the readout is the same with either the sample or standard solution in the light beam. At this point

$$\frac{b_u}{b_s} = \frac{c_s}{c_u}$$

where b_s and b_u are the standard and sample cell lengths and c_s and c_u are the standard and sample concentrations. With a knowledge of the ratio of cell lengths and the concentration of standard, the unknown concentration is easily determined.

The ratio of cell lengths is obtained by using standard solutions in both cells and adjusting their concentrations until the same readout is obtained with either solution in the light beam. The ratio is calculated from the above equation. The ratio cannot be calculated by placing the same solution in the two cells and taking the ratio of the readouts.

In practice, it was easier to take readings of % T on either side of the set point. The plot of % T *vs.* volume is sufficiently linear to obtain the titrant volume at set point by drawing a straight line through the plotted readings.

EXPERIMENTAL

Primary standard potassium dichromate was dissolved in 1M reagent grade sulfuric acid to obtain three stock solutions of 1002.6, 351.0, and 100.26 mg. of chromium per liter. Lingane and Collat (*1*) give the molar absorptivity of dichromate in 1M H_2SO_4 as 369 and state that at this acid concentration the absorbance varies very little with pH.

The instrument used was an unmodified Beckman Model B spectrophotometer equipped with two plexiglass open-topped cells of about 5-cm. cell length and 40-ml. capacity. These cells were held firmly in place from above on the regular cell carriage with an H-shaped bracket fastened by a screw to the carriage.

One molar sulfuric acid was pipetted in 25.00-ml. quantities into both cells. A known amount of dichromate stock solution was added to one cell, which served as the sample cell. The instrument was adjusted as previously de-

[1] Present address, Department of Chemistry, University of Arizona, Tucson, Ariz.

Table I. Cell Length Ratio Measured over Useful Concentration Range of Cr as $Cr_2O_7^{-2}$

b_u/b_s	[Cr] in sample cell in mg./liter
0.9965	137.66
0.9995	123.28
0.9991	107.04
1.0007	91.245
0.9986	74.267
0.9990	57.266
0.9987	57.001
0.9968	53.492
0.9983	48.351
0.9985	43.159
0.9971	38.932
0.9991	37.596
0.9967	31.712
0.9968	25.952
0.9975	20.055
0.9975	16.369
0.9998	15.305
0.9989	13.850
0.9990	13.500
0.9982	12.353
0.9979	10.787
Av. 0.9983	

scribed with this cell in the light beam. Stock solution was then added to the other cell from a 5-ml. microburet, and % T readings were taken when the concentration of the standard cell approached and passed that of the sample cell. A graph was drawn (see Figure 1) and the concentration in the standard cell was determined when the readouts were the same for both cells.

Under the conditions employed the % T reading for the sample (set point) tended to drift. The value of the set point had to be checked often and slight adjustments were made to keep it constant.

RESULTS AND CONCLUSIONS

The ratio of b_u/b_s was calculated from the data obtained as described over a concentration range of 137.7 to 10.79 mg. Cr per liter (see Table I). This ratio was 0.998 with a standard deviation of ±1.13 p.p.t. and a 95% confidence interval of ½ p.p.t. Using the two cells and the method described it is possible to determine dichromate in the concentration range 10^{-3} to $10^{-4}M$ to an accuracy of better than 2 p.p.t.

For substances with molar absorptivities greater than 10^4, this method would allow determinations to an accuracy of ±2 p.p.t. to concentrations of $10^{-5}M$ with an unmodified Beckman Model B spectrophotometer, provided a suitable standard solution was available. The method provides maximum scale expansion for every sample used, requires only one stock solution, and provides a comparison measurement for increased precison. The only disadvantage encountered is the need for the rather bulky equipment and time necessary for the isomation.

Precision and ease of handling could be improved by the use of glass cells instead of the easily scratched and difficult to clean plexiglas cells.

LITERATURE CITED

(1) Lingane, J. J., Collat, J. W., ANAL. CHEM. 22, 166 (1950).
(2) Reilley, C. N., Crawford, C. M., *Ibid.*, 27, 716 (1955).

Magnetic Tape Recording of Analytical Data

Phillip Issenberg,[1] M. L. Bazinet, and Charles Merritt, Jr., Pioneering Research Division, U. S. Army Natick Laboratories, Natick, Mass.

Merritt (*2*) has reported the use of magnetic tape for recording mass spectra derived from a rapid scanning time-of-flight mass spectrometer, and concurrently with this communication the recording of fast scan high resolution mass spectra is reported (*3*). This communication describes the technique employed for recording multiple fast scan mass spectra of compounds separately eluted from a gas chromatographic column and elaborates some of the advantages of magnetic tape in modern analytical data acquisition and processing.

The versatility of magnetic tape is a consequence of the fact that electrical signals are recorded essentially in electrical form. They may be reproduced at some future time at faster, slower, or the same tape speed used for recording the signal. This time scale expansion and compression feature can facilitate interfacing of analytical instruments to data acquisition systems which provide digital records suitable for computer processing. Data recorded in a variety of time frames may be matched, via analog magnetic tape, to a single digitizing system, reducing the complexity and cost of such systems.

Magnetic tape recorders are available with a variety of features, degrees of sophistication, and costs. The specific instrument needed obviously depends on the requirements of the laboratory. Most instrumentation class recorders offer tape speeds of 1⅞, 3¾, 7½, 15, 30, and 60 inches per second (i.p.s.). Tape speed accuracy between 0.2 and 0.5%, and flutter (short term tape speed deviation) between 0.2 and 1.0% are typical of lower price instrumentation recorders. Since recorders may be purchased with seven to fourteen available data channels, it is usually feasible to utilize one channel for recording an accurate timing signal. Oscillators or pulse generators of any frequency and degree of accuracy may be employed depending upon the timing accuracy required in analysis of the data. Thus, an internal standard is provided which makes time measurements essentially independent of tape speed. Independent variables other than time, such as magnetic field strength, voltage, or temperature may also be employed in this manner.

Full scale signal to noise ratios of 40 db. (100:1) are typical of instrumentation recorders operating in the FM mode. This limitation on the dynamic range of the recording system is not serious if the multiple track recording capability is utilized with each track receiving a signal at different attenuation or gain level in a manner similar to that employed with mass spectrometer oscillographic recorders. High speed automatic attenuators may be used to extend the dynamic range for single channel recording.

Magnetic tape recording alleviates many of the data handling problems associated with mass spectrometric analysis of gas chromatographic effluents. If one wishes to identify the maximum number of components of a complex mixture of volatile compounds, a large number of spectra—e.g. 200 to 300—must be recorded during the chromatographic separation. Often many preliminary preparation steps are required to obtain a suitable sample; therefore, it is desirable to extract as much information as possible from a single sample. The probability of missing a significant compound should be minimized. These considerations require either careful judgment by the operator as to when to record spectra during elution of chromatographic peaks, or continuous scanning of spectra during the entire course of elution. If magnetic tape is used as a recording medium, repetitive mass spectral scanning may be employed and all data are permanently recorded in convenient form. The tape may be played back and spectra may be examined carefully prior to permanent recording in any desired format. Data may be examined using an oscilloscope triggered from a scan start pulse recorded on one tape channel, or a recording oscillograph at a slow chart speed may be used.

[1] Present address, Department of Nutrition and Food Science, Massachusetts Institute of Technology, Cambridge, Mass.