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U.S. DEPARTMENT OF COMMERCE/National Bureau of Standards

Standard Reference Materials:

Accuracy in Analytical Spectrophotometry

Standard Reference Materials:

Accuracy in Analytical Spectrophotometry

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PREFACE

As mandated in 1901 by an act of Congress, a prime function of the National Bureau of Standards (NBS) is to develop and maintain the capability for making accurate physical and chemical measurements on the various materials used in science and industry. One of the principal ways in which this mandate has been fulfilled is through the issuance of certified Standard Reference Materials (SRM's). These standards are used to transfer the accurate measurements made at NBS to the user and are frequently issued with detailed publications describing how these SRM's were produced and should be utilized. An NBS series of papers, of which this publication is a member, called the NBS Special Publication – 260 Series, is used for this purpose.

Because of a demonstrated need for more practical transmittance standards, particularly for use in clinical chemistry, a program was initiated in 1969 to study the fundamental principles of optical transmittance and to develop the instrumentation necessary to insure its accurate measurement. This publication is a summary of the work undertaken during the last decade for achieving this objective. It contains a description of the instrumentation that was developed in the NBS Center for Analytical Chemistry for providing accurate transmittance measurements on solid and liquid samples, a summary of the SRM's that were produced and, in addition, brief descriptions of techniques and methodologies that were developed for the accurate chemical characterization of various materials by molecular absorption spectrometry. The scientific papers published in this field are also reviewed together with considerations for future developments.

Inquiries concerning the technical content of this paper should be directed to the authors. Other questions concerned with the availability, delivery, price, and so forth will receive prompt attention from:

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FOREWORD

This work presents a discussion concerning the use of spectrophotometric[1] techniques for the assessment of the optical transmittance of solid or liquid materials, and for the determination of several selected chemical species in a number of matrices. Particular emphasis is placed on the accuracy of the measurements obtained by using the instrumentation, the Standard Reference Materials, and the analytical procedures developed during the last decade in the Center for Analytical Chemistry of the National Bureau of Standards (NBS). This publication is divided into two parts: the first part contains a concise description of the high-accuracy spectrophotometer constructed at the NBS Center for Analytical Chemistry, together with a review of the Standard Reference Materials (SRM's) produced at NBS in the field of spectrophotometry. The second part is concerned with the description of several selected analytical applications of spectrophotometric techniques to the determination of antimony, arsenic, nickel, cobalt, and copper, in various matrices and with a known accuracy.

Future developments in spectrophotometry are considered in the concluding remarks to this work. It includes the potential development of new analytical methods based on the formation of ternary complexes and the use of high performance liquid chromatography as a separation technique for simultaneous multielement analysis.

For further information and convenience, a number of selected publications, directly related to the subjects discussed in this work, are reproduced in the Appendix, together with copies of the certificates for the Standard Reference Materials produced by the National Bureau of Standards in the field of spectrophotometry.

The material used in this work was taken from two lectures delivered by the authors, one at the Society for Applied Spectroscopy meeting in Chicago, Illinois on October 21-22, 1981, with the title "Standard Reference Materials for Spectrophotometry" and the other at the American Society for Testing and Materials Symposium, Boston, Massachusetts, June 27, 1978. This last lecture is published in an ASTM Special Technical Publication 708, pages 45-65, 1980, with the title "Assessment of the Accuracy of Spectrophotometric Measurements and Methodologies".

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[1] Note: the terminology used throughout this publication follows the nomenclature defined by K. D. Mielenz [1][2].

[2] Figures in brackets indicate the literature references at the end of this paper.

ABSTRACT

This publication describes activities undertaken since 1969 within the Center for Analytical Chemistry of the National Bureau of Standards (NBS) in the field of high-accuracy spectrophotometry. The first part of this work presents a summary of the Standard Reference Materials (SRM's) that have been developed for checking the proper functioning of ultraviolet and visible spectrophotometers and includes a description of the high-accuracy spectrophotometer specially constructed in the Center for Analytical Chemistry and subsequently used for performing all of the transmittance measurements. The second part of this publication is devoted to a critical discussion of the analytical factors that can affect the accuracy of selected spectrophotometric procedures that have been widely used at NBS in the characterization of various SRM's.

Key Words: absorbance; accuracy; antimony; arsenic; cobalt; comparative measurements; copper; high-accuracy spectrophotometry; nickel; polarization; scatter; spectral bandpass; spectrophotometric determinations; Standard Reference Materials; stray light; transmittance; transmittance stability; wavelength accuracy.

DISCLAIMER

In order to describe materials and experimental procedures adequately, it was occasionally necessary to identify commerical products by the manufacturer's name or label. In no instances does such identification imply endorsement by the National Bureau of Standards nor does it imply that the particular products or equipment are necessarily the best available for that purpose.

PART ONE

1. General

Ultraviolet and visible spectrophotometry is a measuring technique widely used in science and technology for the determination of numerous chemical elements in a large variety of materials. In the field of health alone, over 95 percent of all quantitative analyses are performed by spectrophotometry and this percentage represents over 3,000,000 daily tests carried out in the chemical laboratories in this country. The need for precision, and particularly for accuracy is of vital importance to these measurements. All determinations that use spectrophotometry are based on the measurement of the optical transmittance of the analytical sample, and the accurate measurement of this parameter is an essential requirement, especially when used not only to establish the optical characteristics of the material, such as the molar absorptivity, but also in enzyme studies, and dosimetry. Transmittance accuracy is also indispensable for interlaboratory exchange and comparison of analytical data.

As indicated by Royden Rand in a comprehensive review [2], numerous interlaboratory studies, using various conventional instruments and various standard solutions, have proven that the accuracy of wavelength and transmittance measurements may be "exceedingly poor". As a result of this situation and following the numerous requests from the users of spectrophotometers, and in particular, from the clinical chemists, the National Bureau of Standards initiated in 1969 a project to study the various factors that affect the accuracy of spectrophotometric measurements, and to provide means to identify, assess, monitor, and, if possible, decrease the uncertainty of such measurements [3].

The sources of variance that are responsible for the uncertainty of spectrophotometric measurements were identified as wavelength accuracy, spectral bandpass, radiation pathlength, stray radiant energy, and transmittance scale accuracy. Since no adequate instrumentation was available to study these parameters individually, in a quantitative manner, and with a proven accuracy, it was decided to design and construct a special high-accuracy spectrophotometer for this purpose [4]. This research instrument was completed and tested in 1970 and was used, since then, for the certification of all the Standard Reference Materials for transmittance and stray radiant energy produced to date at NBS. A condensed description of this instrument is given in the following section.

2. Experimental

2.1. Description of the High-Accuracy Spectrophotometer

The high-accuracy spectrophotometer developed at NBS is a single beam instrument and is illustrated in Figure 1. Its major components described only briefly here are: (a) a radiation source; (b) a monochromator; (c) a sample holder; (d) a system to verify the accuracy of the transmittance measurements; (e) an integrating sphere-photomultiplier unit connected to a digital voltmeter; and (f) a data acquisition and display system. A circular neutral wedge is located between the radiation source and the entrance slit of the monochromator and it is used to select various levels of radiation flux intensities required by the measurements. Adequate quartz-lithium fluoride achromats are used as objectives to produce the necessary beam geometries.

The radiation source for the spectral range 400-1000 nm is a ribbon tungsten filament lamp supplied by a constant current source. The stability of this current is monitored with a potentiometer capable of detecting changes of 2-3 parts in 10^6. For the ultraviolet region from 200 to 400 nm, a deuterium discharge lamp is used. These two light sources are interchangeable and their images can be directed on the entrance slit of the monochromator, using a 45° mirror.

The monochromator is a 1 m Czerny-Turner grating instrument provided with a quartz prism predisperser, functioning as a double monochromator to reduce stray radiant energy. These parts are illustrated in Figure 2.

The sample carrying system provides for manual measurements of one sample and its blank or of seven samples and eight blanks in a sequential order. They can be operated manually or automatically through a microcomputer. The integrating sphere-photomultiplier unit is placed at the end of the optical path and is made from a hollow aluminum sphere provided with a target coated on the inside with $BaSO_4$. The image of the exit slit of the monochromator is projected on the target (Figure 3).

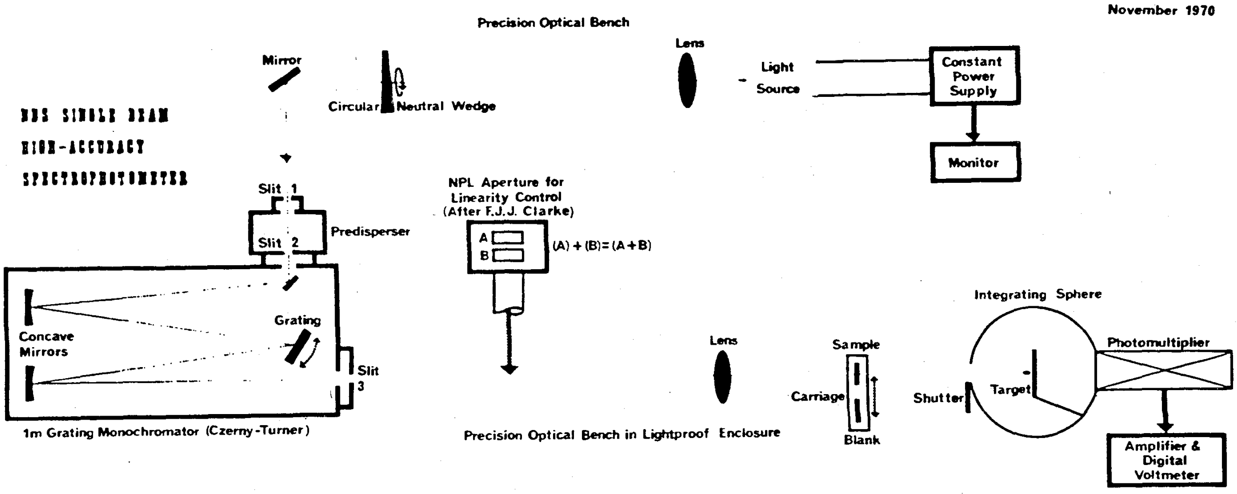

Figure 1. Schematic description of the single beam high-accuracy spectrophotometer. The double aperture unit is placed on the optical bench only when linearity measurements are performed [3,4].

The data acquisition and presentation system consists of a digital voltmeter capable of taking 40 readings per second. The radiation intensities, I, obtained for the sample, and I_o, for the reference beam, passing through air ($I_{o\ air}$) or the blank sample ($I_{o\ blank}$) are used by the microcomputer to calculate transmittance $T\left(\frac{I}{I_{o\ air}}\right)$ or internal transmittance $T_i\left(\frac{I}{I_{o\ blank}}\right)$, which can be expressed as transmittance density ($-\log_{10} T$) or absorbance ($-\log_{10} T_i$).

The microcomputer is also programed to operate all of the measuring sequences, including wavelength settings, light source intensity adjustments, sample changings, and transmittance measurements. These parts are shown in Figure 4.

An essential element of this instrumentation is the system for verifying the accuracy of the transmittance measurements. As mentioned previously, the radiation intensities passing through the sample and reference channels generate corresponding photocurrents i and i_o at the photomultiplier. These photocurrents are substituted for I and I_o in the above transmittance expression and their ratio is the optical transmittance value for the material measured. If these photocurrents can be measured accurately, it follows implicitly that the optical transmittance value will be accurate provided wavelength, radiation pathlength accuracy, adequate spectral bandpass, and freedom of stray radiant energy have been achieved.

A necessary relationship that is implicit for the accurate measurement of photocurrents is that a linear relation exists between the radiation flux intensity and the corresponding photocurrent generated by the photodetector.

Various methods may be used to measure the linearity of these photocurrents. A technique based on the radiation addition principle using two apertures was selected for its simplicity and freedom from errors. The two aperture system, illustrated in Figure 5, consists of a metal plate provided with two rectangular windows A and B. Each window can be closed by shutters operated remotely through pneumatic controls [5]. This aperture system is placed following the exit slit of the monochromator and always within the optical solid angle of the instrument. The image of the apertures is then produced on the target of the integrating sphere. The linearity verification consists of measuring the photocurrent produced with aperture A open and B closed; then B open and A closed; and finally with both apertures A and B open. If the system is linear, the sum of the photocurrents obtained separately with aperture A and B should be equal to the photocurrent obtained when both apertures A and B are open. If this is not the case, the system shows a nonlinearity which is proportional to the amount by which the sum of (A) + (B) differs from (A + B). This difference is then used to correct the transmittance measurements. During the individual measurement of (A), (B), and the combined (A + B), experimental conditions should remain identical.

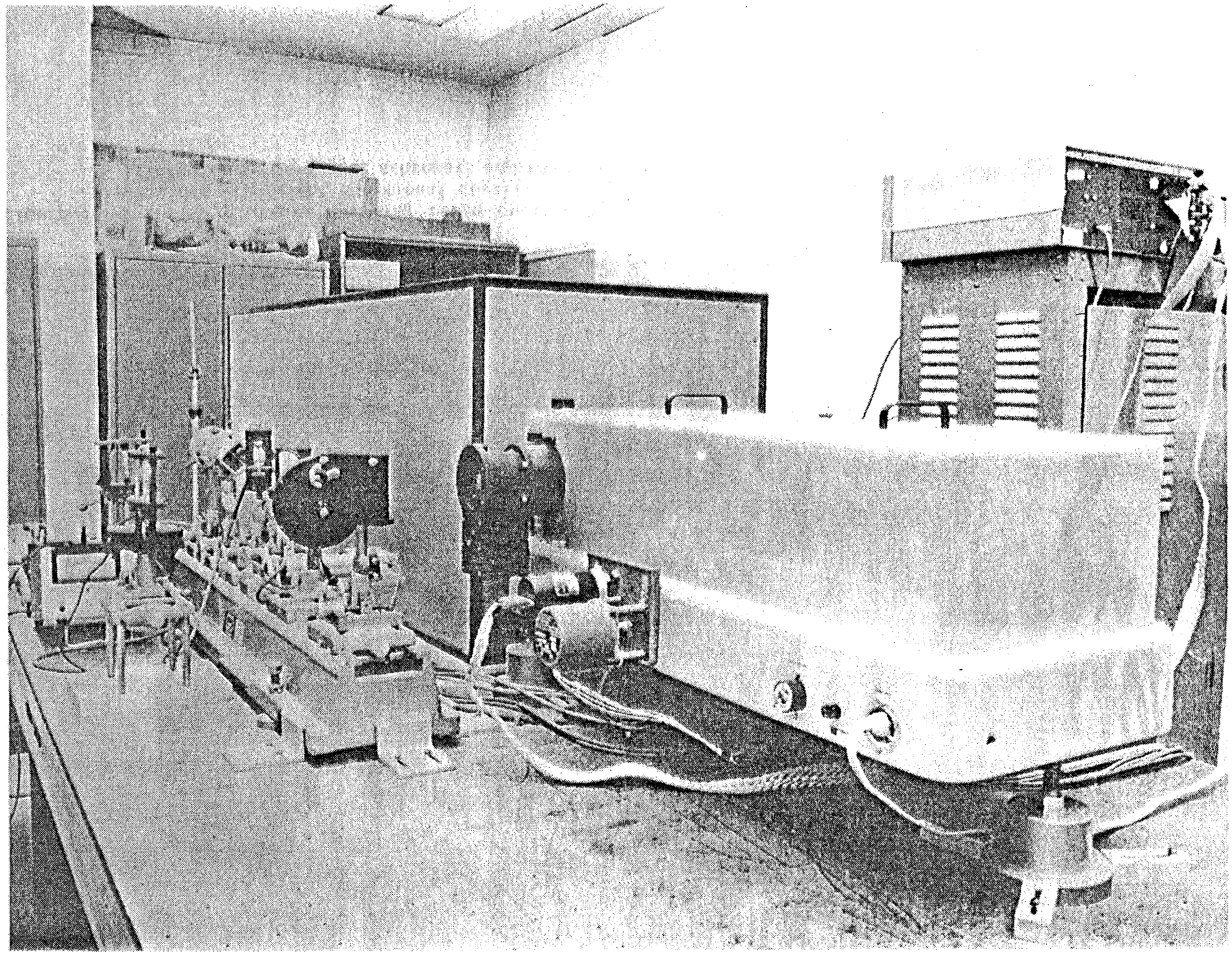

Figure 2. Arrangement of the two radiation sources used with the high-accuracy spectrophotometer. A low power laser beam is used to verify the optical alignment. Left: the tungsten filament lamp for measurements in the visible. On the optical bench: the deuterium low pressure discharge source for measurements in the ultraviolet. Also on the optical bench: the 45° mirrors and the circular neutral wedge. At right: the 1 m Czerny-Turner spectrometer provided with the predisperser, stepping motor and encoder. The sample holder, integrating sphere-photomultiplier unit, and the two achromat objectives are placed on an optical bench located in the light tight enclosure visible at the rear. Also at right, the console carrying the electronic and pneumatic controls described in Figure 4.

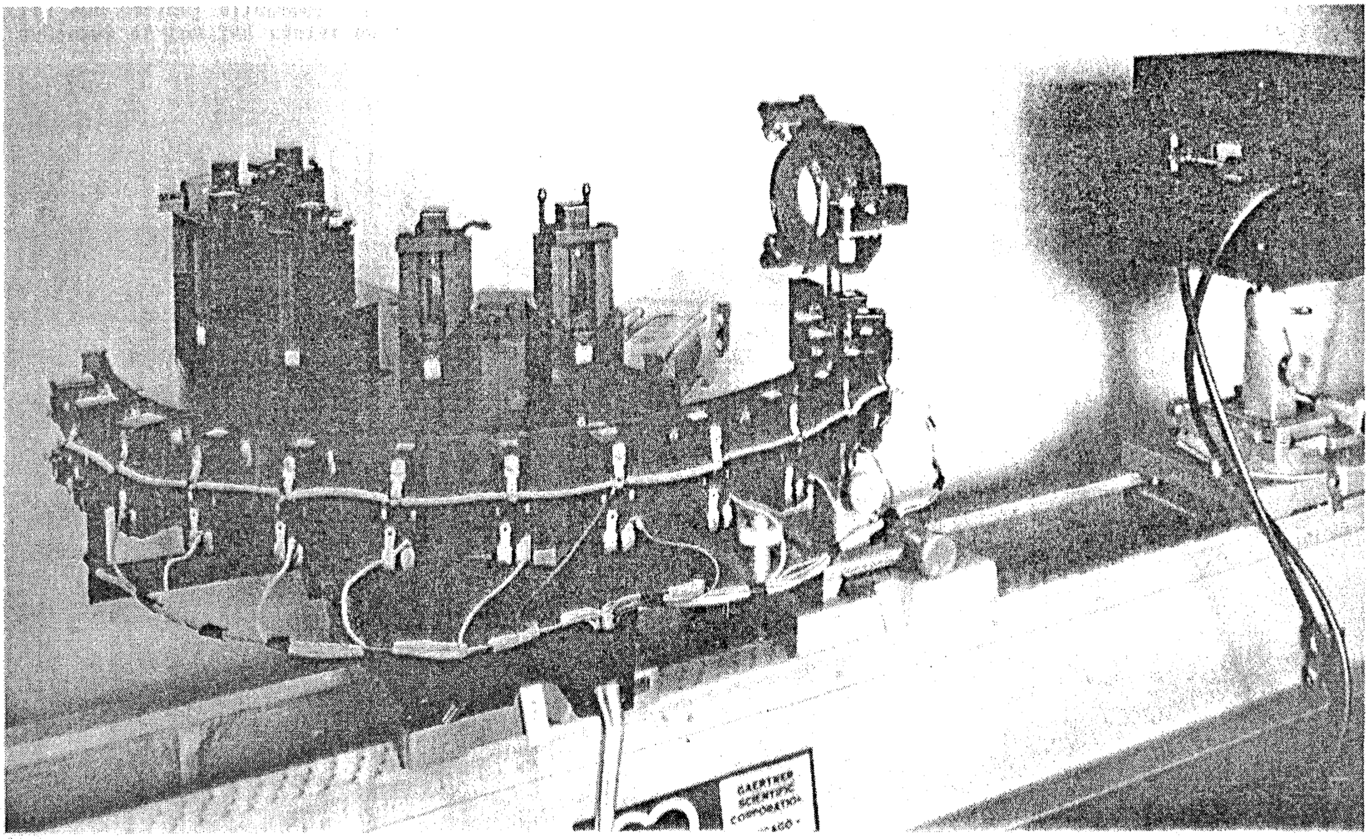

Figure 3. Circular platform carrying seven cuvette or filter holders. An improved unit (not illustrated) is provided with eight additional cuvette or filter holders for the blank samples. At right: one of the two lithium fluoride-quartz achromats and the integrating sphere-photomultiplier unit.

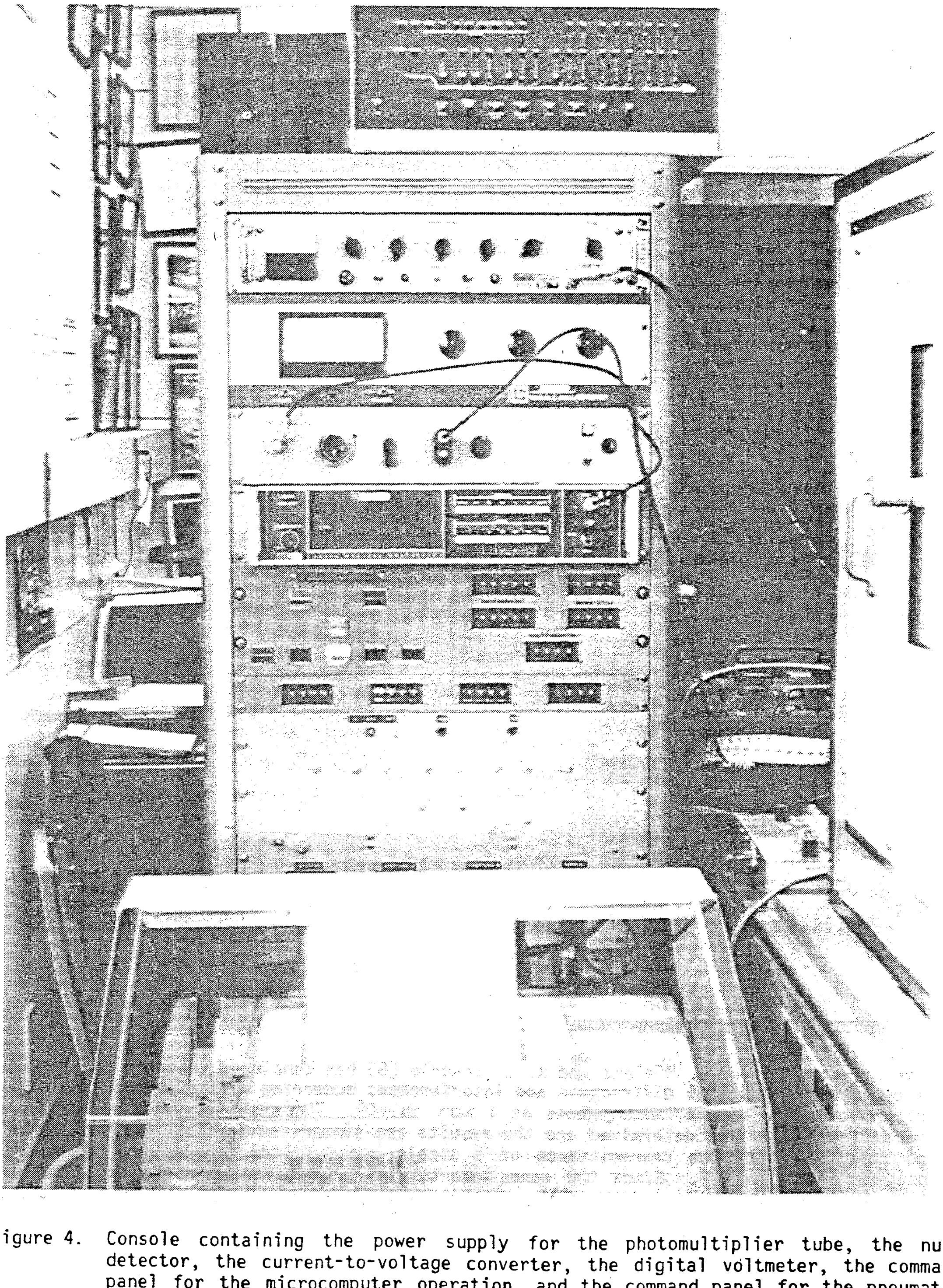

Figure 4. Console containing the power supply for the photomultiplier tube, the null detector, the current-to-voltage converter, the digital voltmeter, the command panel for the microcomputer operation, and the command panel for the pneumatic controls. The electric switches for operating the circular neutral wedge are also located on this panel. Middle right: potentiometer for monitoring the dc current supplied to the tungsten ribbon filament lamp. At the bottom left: teletype for data presentation. Right: light panel indicating the position of the sample holder. On top: microcomputer.

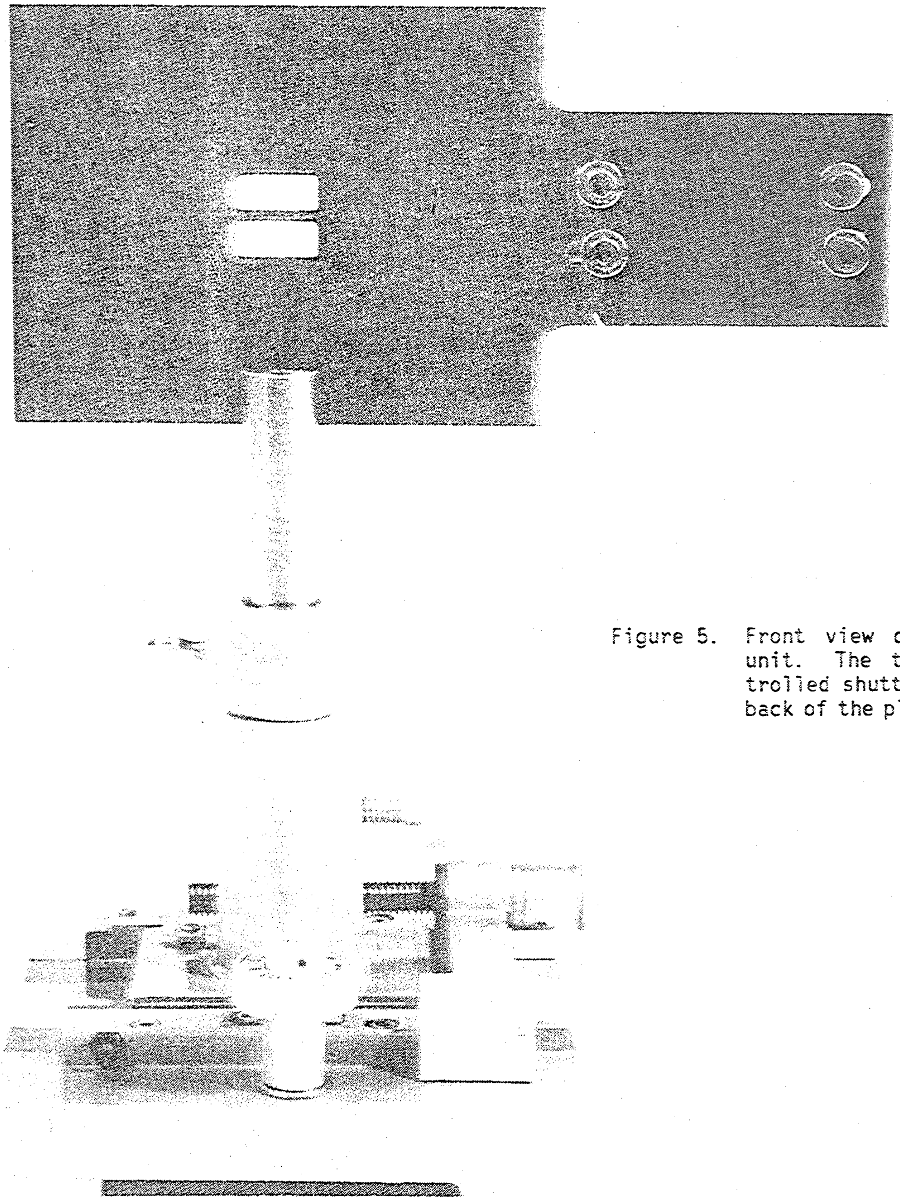

Figure 5. Front view of the double-aperture unit. The two pneumatically controlled shutters are located on the back of the plate.

A critical study by K. D. Mielenz and K. L. Eckerle [6] has concluded that the possible causes of errors generated by optical diffraction and interferences occurring at the apertures system will not affect the accuracy of the measurements at 1 part in 10^5. The stability of the various electronic and optical parts was determined and the results are summarized in Table 1. From these data it can be concluded that the transmittance of a stable glass filter can be determined with an uncertainty of 1 part in 10^4. Since the same uncertainty is obtained when the linearity of the photocurrent is measured, the accuracy of the transmittance is also assessed with an uncertainty of 1 part in 10^4.

A similar high-accuracy spectrophotometer, designed by K. D. Mielenz et al. [7], was constructed and is used in the NBS Center for Radiation Research for the study of various physical parameters in spectrophotometry. This instrument differs from the one described here in that it uses reflecting rather than refracting optics. Some of the parameters studied on this research instrument are: interreflection phenomena, polarization, homogeneity of the photomultiplier sensitive surface, effectiveness of integrating spheres and diffusers, scatter and homogeneity of transparent solid optical filters, wavelength calibration of glass filters, etc., [8].

Table 1. Summary of Functioning Characteristics of the High-Accuracy Spectrophotometer.

- Stability of the electronic system	1 to 2 parts in 10^5
- Stability of the electronics and PM tube (dark)	4 parts in 10^5
- Stability of electronics, PM tube, and radiofluorescent source	7 parts in 10^5
- Stability of electronics, PM tube, and tungsten ribbon filament	2.2 parts in 10^4
- Reproducibility of transmittance measurements	1 part in 10^4

2.2. Standard Reference Materials

The high-accuracy spectrophotometer housed in the Center for Analytical Chemistry is described in this paper because it represents the primary transmittance standard. The only way known to us by which the accurate transmittance values determined with this instrument, on solid and liquid materials in the spectral range from 200-800 nm, can be transmitted to the users of spectrophotometers is with the help of Standard Reference Materials, or SRM's. These SRM's are defined as "well-characterized materials produced in quantity and certified for one or more physical or chemical properties", and their objective is to assure the accuracy and compatibility of measurements throughout the nation, by transmitting to the user the accurate values obtained at NBS.

A number of SRM's have been prepared at NBS for the verification of the accuracy of the wavelength and transmittance scales of conventional spectrophotometers, and for assessing the stray radiant energy. Cuvettes having a known radiation pathlength and a fluorescent SRM have also been issued. These SRM's are described in Table 2 which presents a summary of their major characteristics together with information on the publications prepared at NBS that describe in detail the selection, characteristics, preparation, calibration procedures, and uses of these materials.

2.2.1. SRM 930

The first SRM for spectrophotometry, issued by NBS in March 1971, is the transmittance standard illustrated in Figure 6. It consists of three glass filters made of Schott NG-4 and NG-5 optically neutral glasses cut to fit a metal holder that can be placed in the cuvette compartment of conventional spectrophotometers. Their nominal transmittances, in the visible spectral range from 400-635 nm, are 10, 20, and 30 percent. Each transmittance is certified with an uncertainty of 0.5 percent, at the time of certification, at wavelengths of 440, 465, 546, 590, and 635 nm and with spectral bandpasses of 2.2-6.0 nm according to the wavelength.

Each metal holder is provided with two shutters for the protection of the glass, and a fourth empty holder is provided, as shown in Figure 7, for use in the reference beam. The filters are placed in a black anodized aluminum holder for storage and transportation. Before certification, each glass filter is examined using a stereomicroscope (x30) for surface defects. It is then numbered for identification, cleaned in an ultrasonic bath, placed in a stainless steel holder, and finally cleaned in a Soxhlet extractor with isopropyl alcohol. The clean filters are then mounted in the metal holders. The equipment is shown in Figure 8, and the extractors are illustrated in Figure 9. All operations are performed in a dust-free polypropylene hood with vertical laminar air flow.

The cleaning process described here is satisfactory for the purpose of this work. A detailed bibliography of the numerous works on this problem is given by P. Bruce Adams [9]. In addition a chapter on "The Cleaning of Glass" is part of the book by L. Holland "The Properties of Glass Surfaces" [10], where this subject is discussed in depth.

Transmittance measurements are first performed on these filters after waiting several weeks for their freshly-cleaned glass surfaces to "stabilize" and then also six months later to verify the optical stability of the material. The filters are issued when these two sets of measurements agree within 0.3 percent. As discussed later, this aging period is an essential condition to insure that SRM 930 has the transmittance stability required.

Table 2. Standard Reference Materials for Spectrophotometry Provided by the National Bureau of Standards.

SRM	Material	Composition
Solid Filters		
930 Glass Filters for Spectrophotometry	Solid filters made of Schott NG 4 and NG 5 optically neutral glass. Obtained from the Schott-Jenaer Glaswerk, Mainz, Germany	Three filters with nominal transmittances of 10, 20, 30%. Each filter is mounted in a black anodized aluminum holder provided with front and rear shutters. The filters are stored in a cylindrical aluminum container.
2030 Glass Filter for Transmittance Measurements	Solid filter made of Schott NG 5 optically neutral glass. Obtained from the Schott-Jenaer Glaswerk, Mainz, Germany,	One glass filter with a nominal transmittance of 30% in a black anodized aluminum holder with shutters. The filter is stored in a wood-like plastic box.
2031 Metal-on-Quartz Filters for Spectrophotometry	Solid filters made of semi-transparent evaporated chromium-on-fused silica (non-fluorescent) plates.	Three filters with nominal transmittances of 10, 30, 90%. Each filter is made from two fused silica plates assembled by optical contact. The 90% filter is made by assembling 2 clear plates. The 10% and 30% filters are made by assembling a fused silica plate which carries a film of semitransparent chromium to produce the desired transmittance, and a clear plate. Each filter assembly is placed in a metal holder provided with shutters.
Liquids and Cuvettes		
931b Liquid Absorbance Standards for Ultraviolet and Visible Spectrophotometry	Liquid filters made from a solution of Co and Ni metals dissolved in a mixture of nitric-perchloric acids. The pH of the solution is about 1. Prepared in the NBS Center for Analytical Chemistry.	Three solutions and a blank are delivered in sealed ampoules and have nominal absorbances of 0.1 to 0.9.
935 Crystalline Potassium Dichromate for Use as an Ultraviolet Absorbance Standard	Crystalline potassium dichromate of established purity obtained from the J. T. Baker Chemical Co., Phillipsburg, New Jersey.	The crystalline material of reagent grade purity is offered in glass bottles provided with plastic screw caps.

Certification	Use	Remarks
The transmittance of each filter is measured with the high-accuracy spectrophotometer at λ 440, 465, 546.1, 590, 635 nm, using spectral bandpasses of 2.2, 2.7, 6.5, 5.4, 6.0 nm respectively.	This SRM is intended as a reference source for the verification of the transmittance and absorbance scales of conventional spectrophotometers.	The use of this SRM is limited to the visible spectrum and requires narrow spectral bandpasses. For further details, consult the Certificate included in the Appendix; see also NBS Special Publication 260-51.
The transmittance of the filter is certified as for SRM 930 but only for λ 465 nm and for a nominal transmittance of 30%.	This SRM is intended as a reference source for one-point verification of the transmittance or absorbance scales of spectrophotometers.	Same as for SRM 930. SRM 2030 is particularly useful in connection with the chemical analysis of spices. A copy of the Certificate is included in the Appendix.
The transmittance of each filter is measured with the high-accuracy spectrophotometer at λ 250, 280, 340, 360, 400, 465, 500, 546.1, 590, 635 nm.	This SRM is intended as a reference source for the verification of the transmittance and absorbance scales of conventional spectrophotometers in the ultraviolet and visible spectral region.	SRM 2031 has good optical neutrality and can be used to calibrate spectrophotometers with wide spectral bandpasses from about 200 to 800 nm (with possible extension to λ=3 μm). For further details, consult the Certificate in the Appendix; see, also NBS Special Publication 260-68.
The absorbance of each solution was measured at λ 302, 395, 512, 678 nm using spectral bandpasses of 1.0, 1.7, 2.0, 6.5 nm respectively. The certification is made with an uncertainty of about ±1.0%.	This SRM is primarily intended for the verification of the accuracy of transmittance or absorbance scales of spectrophotometers.	Requires the use of SRM 932 and of narrow spectral bandpasses. Provides only one certified value in the ultraviolet. For further details consult the Certificate included in the Appendix.
The apparent specific absorbance is certified for five concentrations at λ 235, 257, 313, 345, 350 nm using spectral bandpasses of 1.2, 0.8, 0.8, 0.8, 0.8 nm respectively, using SRM 932.	This SRM is intended to be used as a reference standard for the verification of the accuracy and linearity of the absorbance scale of spectrophotometers.	Requires accurate preparation of solutions of potassium dichromate in 0.001N perchloric acid. Requires the use of SRM 932 and of narrow spectral bandpasses. Can be used only in the ultraviolet. For further details, consult the Certificate included in the Appendix; see also NBS Special Publication 260-54.

Table 2. continued.

SRM	Material	Composition
932 Quartz Cuvette for Spectrophotometry	All-quartz cuvette made of non-fluorescent fused silica of optical quality.	Each cuvette is made of fused silica and is provided with a Teflon stopper. The transparent windows are attached to the body of the cuvette by direct fusion. The cuvettes are stress-relieved by proper annealing. The cuvette is stored in a transparent plastic container.
Stray Light		
2032 Potassium Iodide for use as a Stray Light Standard	Crystalline potassium iodide of established purity, obtained from the J. T. Baker Chemical Co., Phillipsburg, New Jersey.	The reagent-grade, crystalline KI is 99.8% pure as indicated by the manufacturer. Moisture is 0.007%. Homogeneity was determined by absorbance measurements at 265, 267, and 270 nm, and was found adequate.
2033 Potassium Iodide for use as a Stray Light Standard with Radiation Attenuator	Crystalline potassium iodide of established purity, and two semi-transparent evaporated metal-on-fused silica filters contained in a metal holder provided with shutters. The KI was obtained from the J. T. Baker Chemical Co., Phillipsburg, New Jersey. The filters and the holder were made by the NBS Instrument Shop.	Potassium iodide as for SRM 2032. Two semi-transparent evaporated chromium-on-fused silica (non-fluorescent) filters of optical quality.
Wavelength		
2009 Didymium Glass Filter for Checking the Wavelength Scale of Spectrophotometers	The didymium glass was prepared by Corning Glass Works, Corning, New York, as Corning 5120 Filter Glass.	Rare earth oxides in a glass matrix. This filter is 1 cm wide, 3 cm high, and 3 mm thick. Each filter is placed in a metal holder which fits into the cuvette holder of the spectrophotometer.
2010 Didymium Glass Filter for Checking the Wavelength Scale of Spectrophotometers	Same as SRM 2009.	Rare earth oxides in a glass matrix. This filter is 5.1 cm square and 3 mm thick.
2013 Didymium Glass Filter for Checking the Wavelength Scale of Spectrophotometers	Same as SRM 2009.	Same as SRM 2009.

Certification	Use	Remarks
The nominal inner pathlength is 10 mm and it is certified at 10 points along the height of the transparent windows with an uncertainty of ±0.0005 mm. The cuvettes are 12.5 mm square and 48 mm high.	To be used when accurate transmittance or absorbance measurements are made on liquid samples. Should be used in conjunction with SRM's 931, 935, 2032, and 2033.	For further details, consult the Certificate included in the Appendix; see also NBS Special Publication 260-32.
The specific absorbances were measured with the high-accuracy spectrophotometer at 240, 245, 250, 255, 265, 270, and 275 nm and 23.5 °C using a spectral bandpass of 0.2 nm. The measurements were performed using SRM 932 quartz cuvettes.	SRM 2032 is to be used to assess heterochromatic stray light in the ultraviolet region below 260 nm, in absorption spectrophotometers. It is recommended that SRM 932 be used in the measurements.	SRM 2032 should be stored in the original, low-actinic glass bottle and cardboard container, protected from exposure to light and humidity. The estimated stability is 3 years.
Same as for SRM 2032. The transmittance of the evaporated metal-on-fused silica filters was measured at 255 nm with the high-accuracy spectrophotometer. The nominal value is 10% for each filter, and a combined value of 1%.	SRM 2033 is to be used to assess heterochromatic and isochromatic stray light in absorption spectrophotometers.	Same as for SRM 2032. The two semi-transparent evaporated metal-on-fused silica filters in the metal holder should be stored in the plastic container provided with SRM 2033.
The wavelengths of maximum absorption were determined with a high-precision spectrophotometer for bandwidths in the range 1.5 to 10.5 nm and for 14 to 24 wavelengths in the range 400 to 760 nm. The instrument has a wavelength accuracy of 0.04 nm.	The filters are to be used in calibrating the wavelength scale in the visible spectral region for spectrophotometers having nominal bandwidths in the range 1.5 to 10.5 nm.	SRM 2009 was not measured individually. It is recommended for most applications. For further details, consult NBS Special Publication 260-66.
Same as SRM 2009.	Same as SRM 2009.	Same as SRM 2009.
Each filter was measured separately; hence the data provide a more accurate representation of the optical properties of the individual filter.	Same as SRM 2009	SRM 2013 was measured individually and should be used when assurance in the evaluation of an instrument's wavelength scale is highly critical. For further details, consult NBS Special Publication 260-66.

Table 2. continued.

SRM	Material	Composition
2014 Didymium Glass Filter for Checking the Wavelength Scale of Spectrophotometers	Same as SRM 2009	Same as SRM 2009.
2034 Holmium Oxide for use as Wavelength Standard in Spectrophotometry and Fluorescence Spectrometry. (Planned SRM)	Holmium oxide of established purity dissolved in perchloric acid.	Holmium oxide is to be offered in solutions in sealed 10 mm quartz cuvettes.
Fluorescence		
936 Quinine Sulfate Dihydrate	The quinine sulfate dihydrate was a special lot of material obtained from J. T. Baker Chemical Co., Phillipsburg, New Jersey.	SRM 936 contains 1.7% impurities determined by liquid chromatography and believed to be dihydroquinine sulfate dihydrate. The water content is 4.74±0.05% determined by the Karl Fisher method and 4.57±0.04% by weight loss. Theoretical value is 4.60%.

NOTE: For further scientific information concerning the following Standard Reference Materials, write or call at the National Bureau of Standards, Washington, D. C. 20234: SRM's 930; 931; 932; 935; 2030; 2031; 2032; and 2033: Dr. R. W. Burke, Inorganic Analytical Research Division, Chemistry Building, Room B-216. Phone: 301-921-2141. SRM 936: Dr. R. A. Velapoldi, Deputy Director, Center for Analytical Chemistry, Chemistry Building, Room A-311. Phone: 301-921-2852. SRM's 2009; 2010; 2013; and 2014: Mr. K. L. Eckerle, Radiometric Physics Division, Center for Radiation Research, Metrology Building, Room A-321. Phone: 301-921-2791; for Scientific information concerning physical parameters involved in spectrophotometry and luminescence measurements Dr. K. D. Mielenz, Chief, Radiometric Physics Division, Center for Radiation Research, Metrology Building, Room B-304. Phone: 301-921-3864.

For information concerning the availability and purchase of the SRM's discussed in this paper contact: Mr. R. W. Seward, Technical Representative, Office of Standard Reference Materials, Chemistry Building, Room B-313. Phone: 301-921-2045. The NBS Publication discussed in this paper can be purchased from the: Superintendent of Documents, U.S. Government Printing Office, Washington, D. C. 20034. Please write for information concerning availability, delivery, and price.

Certification	Use	Remarks
Same as SRM 2013.	Same as SRM 2009.	Same as SRM 2013.
The wavelengths of maximum absorption of holmium oxide solutions in perchloric acid will be determined for various spectral bandpasses at the nominal wavelengths; 279, 288, 338, 361, 386, 418, 453, 536, 637 nm.	This SRM is to be used as a reference standard for the verification of the accuracy of the wavelength scale of absorption and fluorescence spectrometers, in the ultraviolet and visible spectral regions.	Holmium oxide solutions in perchloric acid were selected as a wavelength standard because the absorption bands are narrower than those of the holmium oxide glass. The use of solutions in conjunction with a diffusing screen (Teflon) placed at 45° in the 10 mm cuvette permits the use of SRM 2034 for the wavelength calibration of fluorescence spectrometers. The issuance of SRM 2034 is projected for the near future.
The material is certified for the relative molecular emission spectrum $E(\lambda)$, in radiometric units for a solution of 1.28×10^{-6} mol/L in 0.105 mol/L perchloric acid, using an excitation wavelength of 347.5 nm. The certified values of the molecular emission spectrum at 5 nm intervals from 375 to 675 nm are given. This certification was made with the NBS reference fluorescence spectrometer.	For evaluation of methods and the calibration of fluorescence spectrometers. A solution of 0.1 mg/mL in 1000 mL 0.105 mol/L perchloric chloric acid is recommended. It should be stored in the dark in a well-stoppered glass bottle. This solution is stable for three months. SRM 936 is for "in vitro" diagnostic use only.	The material should be kept in its original bottle and stored in the dark at 30 °C or less. Under these conditions SRM 936 is stable for three years. See NBS Special Publication 260-64.

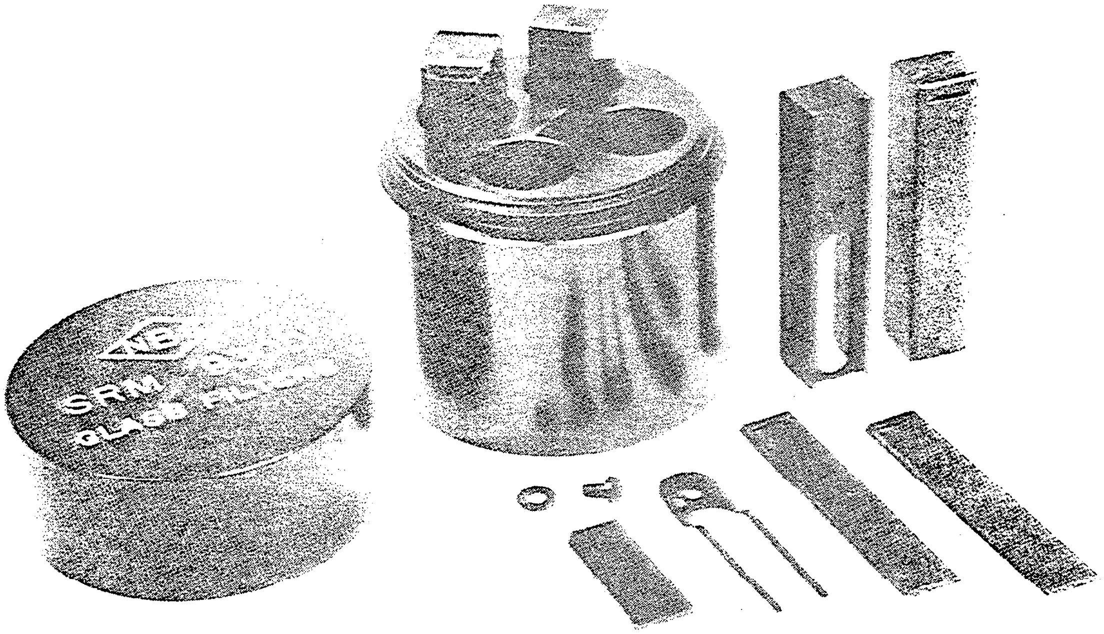

Figure 6. Illustration of SRM 930, Glass Filters for Spectrophotometry, showing one of the three glass filters, the metal holder with its two plastic shutters, and the retaining spring with its nylon screw and washer. A fourth empty filter holder is provided, and the four holders are stored in the cylindrical metal container.

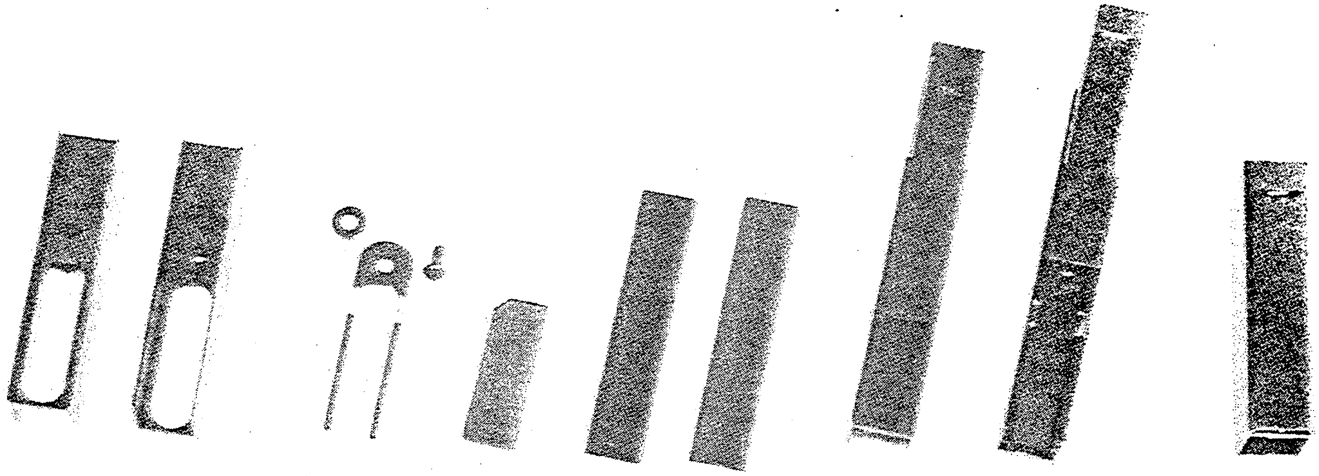

Figure 7. Details of the filter holder. From left to right: front and rear view of the holder body; retaining spring and nylon screw with washer; glass filter; plastic shutters; the holder with the front and rear shutters on; and the filter with both shutters closed.

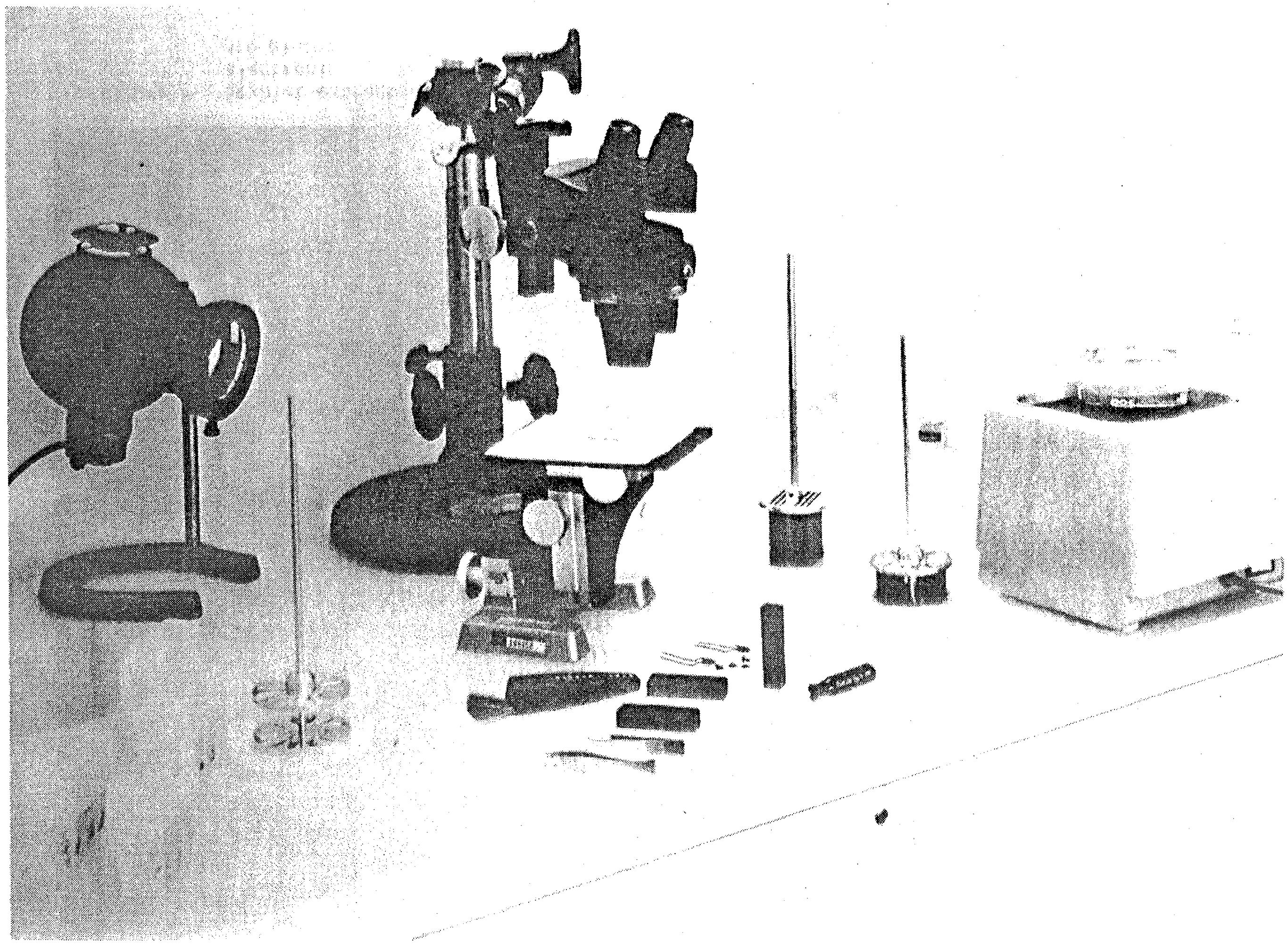

Figure 8. Instrumentation used to examine, clean, and mount the glass filters that constitute SRM 930. The binocular microscope, the ultrasonic cleaner and the various filter holders and tools are placed in an all-polypropylene hood provided with a vertical air flow.

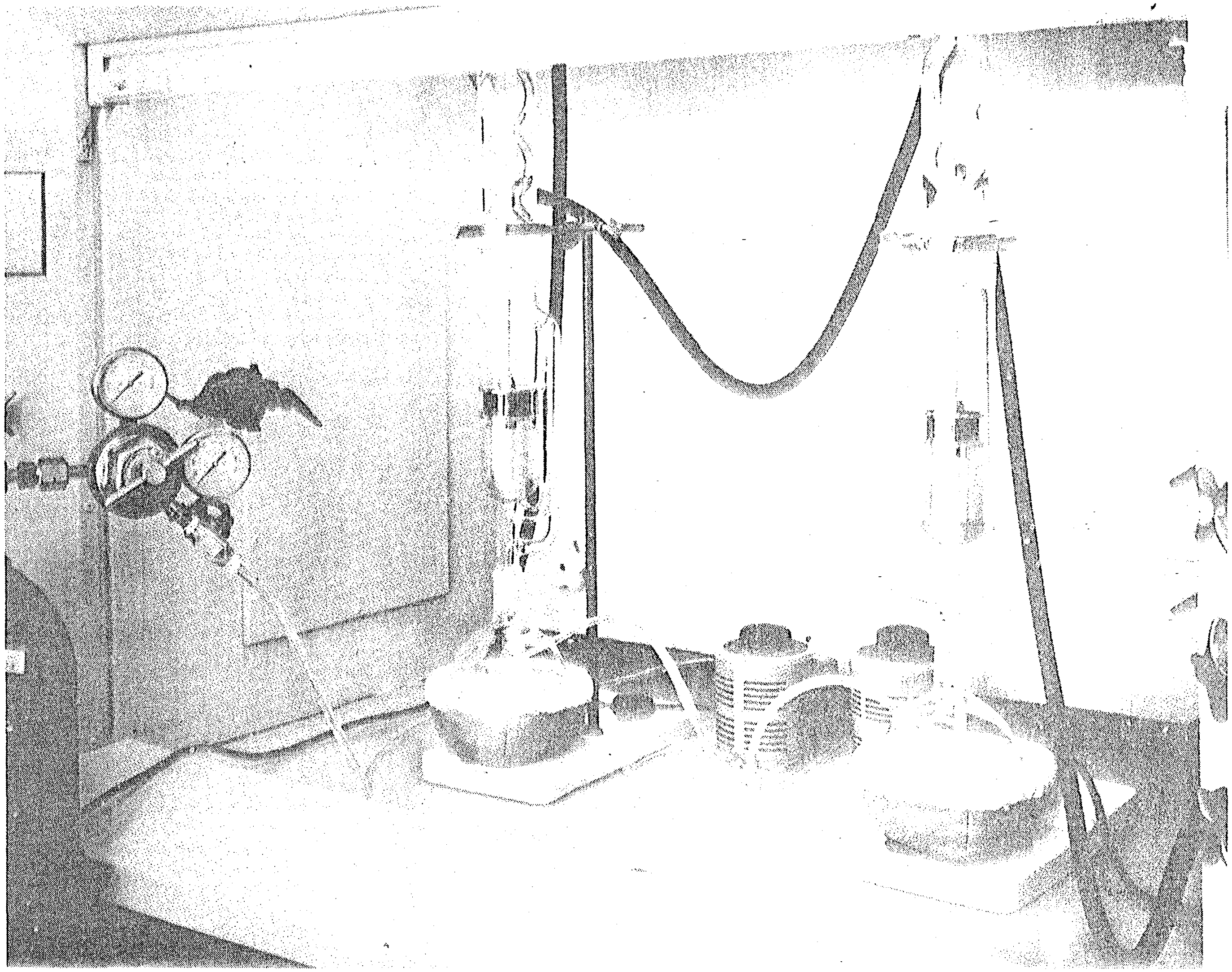

Figure 9. Soxhlet extractors used for the final cleaning of the glass filters, using isopropyl alcohol of electronic grade. Both filter sizes used: 50 x 50 mm and 30 x 10 mm are illustrated. They are placed in stainless steel holders with a capacity of 6 and 12 filters respectively.

2.2.2. SRM 2030

Another glass filter standard is SRM 2030 which is derived from SRM 930. This material, illustrated in Figure 10, is made of only one glass filter calibrated at only one wavelength, namely 465 nm (SBP 2.7 nm). SRM 2030 was issued at the request of the spice industry which uses it as a reference standard in the methods used for determining the quality of various spice extracts. The use of this SRM has decreased the uncertainty of the spectrophotometric analyses from about 10 percent to 2 to 3 percent.

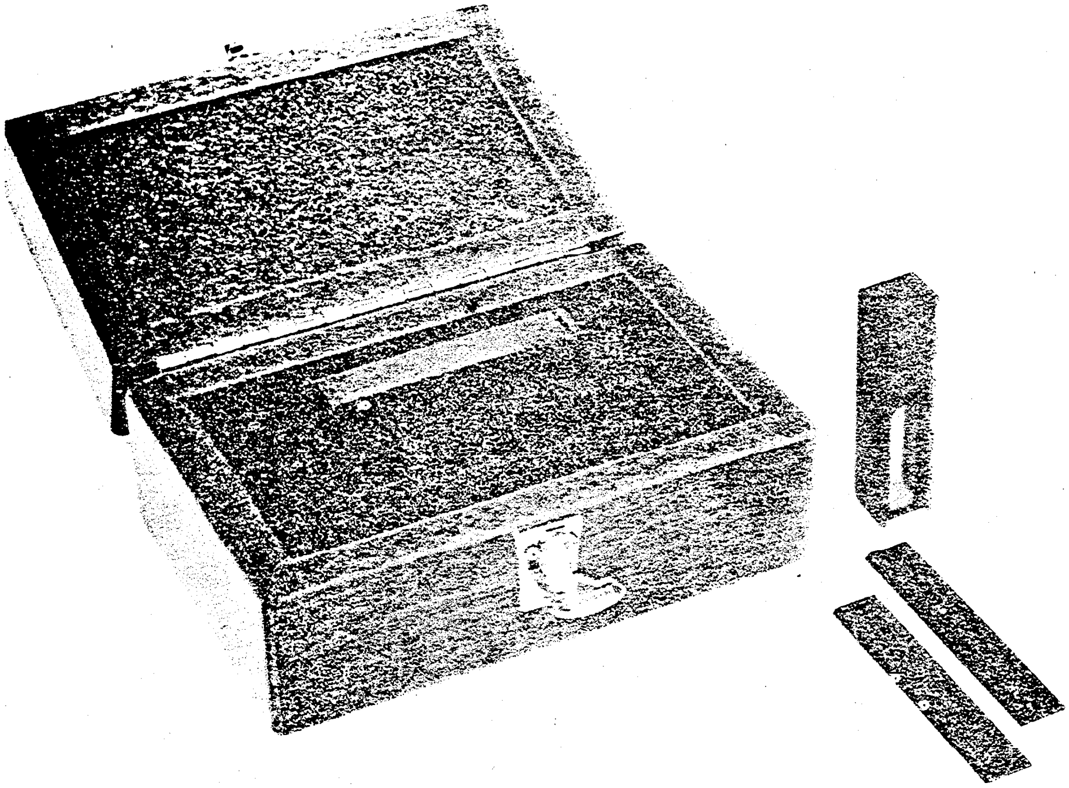

Figure 10. Illustration of SRM 2030 consisting of one glass filter Schott NG-5 in its metal holder provided with the removable front and rear shutters, and an empty filter holder, in the storage box.

2.2.2.1. Monitoring the Stability of Spectrophotometers

The short and long term stability of a spectrophotometer is directly related to its overall design and construction. For many modern commercial instruments, the short term (day-to-day) variation in transmittance (T) does not exceed 5×10^{-4}. This variability corresponds to an absorbance (A) variability of 2×10^{-3} at $T = 0.1$ ($A = 1$) and to a ΔA of 3×10^{-4} at $T = 0.8$ ($A = 0.1$). For a longer term, namely, month-to-month, the spectrophotometer may exhibit results that indicate a gradual shift in performance outside of these limits. This shift is indicative of aging, wear, and deterioration of the instrument's various optical, mechanical, and electronic components and should be monitored periodically. Any abrupt change in the spectrometer's performance indicates a malfunction and corrective action should be taken immediately. Corrective action should also be taken whenever cummulative gradual changes are sufficient that the performance of the instrument is outside of the manufacturers' specifications.

The recommended way to monitor the short and long term stability of a spectrophotometer is by construction of a control chart. The essential requirement for such a control chart is the availability of a stable standard. Of the several types, the most convenient are glass filters, specially selected for their stability. Of the several kinds of glasses that can be used, the SRM 930 type is the most suitable for general purposes. The chart is constructed by making transmittance measurements at several well-spaced wavelengths and plotting the results on the ordinate and the date on which the measurements were made on the abscissa. Measurements should be performed daily. Additional data which should be recorded at the time of measurement are the slit width and the temperature.

A control chart for one of the precision spectrophotometers in the Center for Analytical Chemistry at NBS is shown in Figure 11. The upper plot (a) was obtained from measurements taken at 635.0 nm and the lower plot (b) from measurements taken at 590.0 nm. An examination of these data shows that the stability of this instrument varied during the period of October 22 to February 5 from 32.17 to 32.28 percent T with an average of 32.23 percent T at 635.0 nm. At 590.0 nm the corresponding values were 30.79 and 30.90 percent T with an average of 30.85 percent T. For each wavelength, the spread is 0.11 percent T. This spread results from the inherent instability of the spectrophotometer and of the glass filters used. The performance of this spectrophotometer changed markedly on February 8 when a transmittance of 31.94 and 30.59 were obtained at 635.0 and 590.0 nm, respectively. Since these values were well outside the spread for this instrument, the manufacturer's service representative was consulted. An examination of the spectrophotometer revealed that a potentiometer was malfunctioning and this part was replaced. The spectrophotometer was checked again on February 18 and the transmittance values, measured on the same glass filters, were within the spread previously established for the instrument. The periodic verification of the stability of spectrophotometers as described above and illustrated in Figure 11 should be used in analytical laboratories on a routine basis. A control chart, in conjunction with corresponding slit width data, provides an excellent diagnostic check when an instrument should be serviced or a particular component replaced.

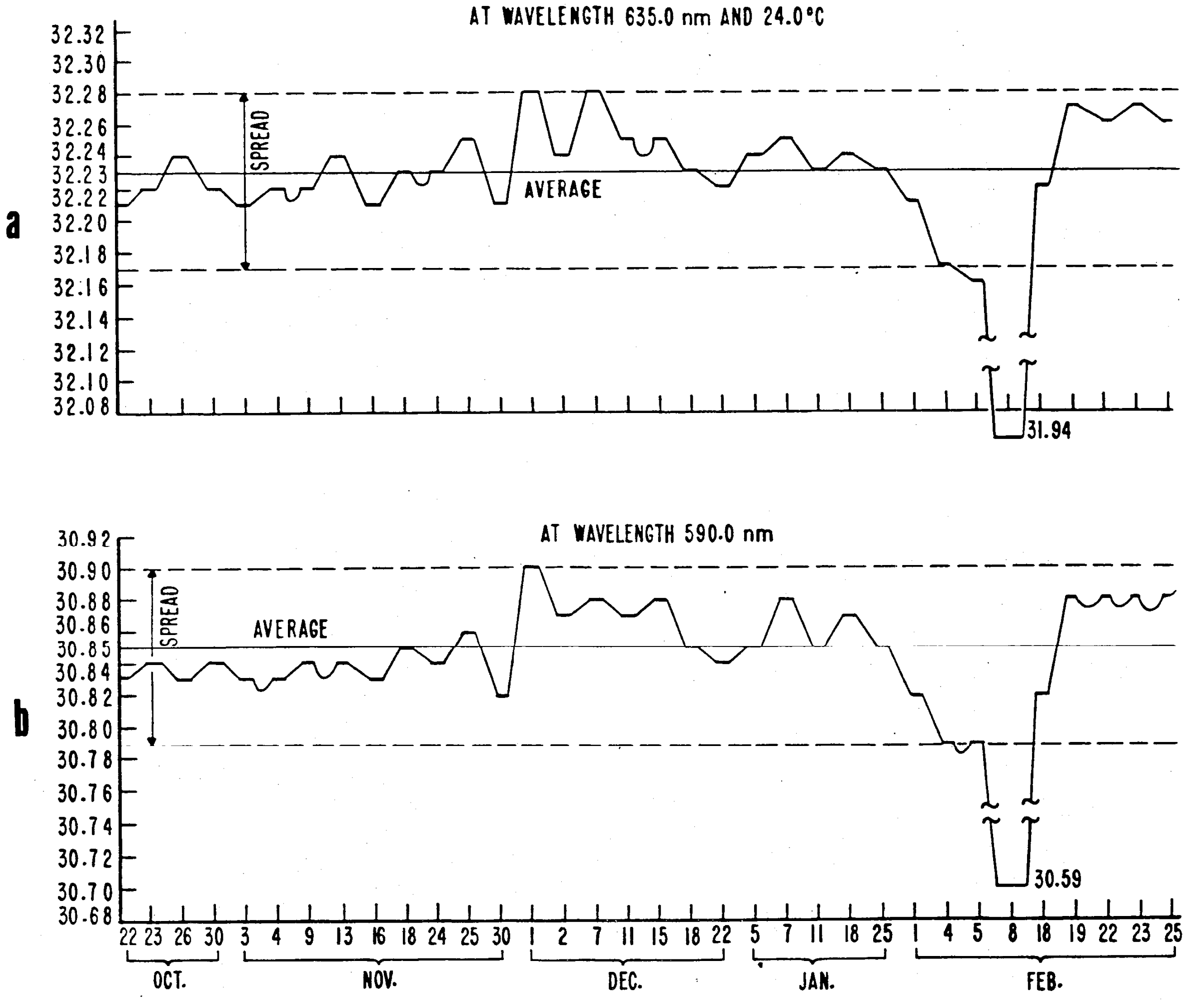

Figure 11. Control chart for a precision spectrophotometer showing the variation of transmittance with time for a Schott NG-5 neutral glass filter at 635.0 nm (a) and 590.0 nm (b).

2.2.2.2. Stability of Glass Filters

Of the many characteristics of these glass filters, only two will be mentioned here. Briefly, these are the stability of the transmittance and the capability of these transmittance measurements to be transmitted to other laboratories. These properties are obviously of major significance not only in relation to these standards, but for any type of transfer standards.

Several sets of SRM 930 were tested over a period of 10 years, during which transmittance measurements were performed at selected time intervals and at four to five wavelengths in the visible spectral range. The data from Table 3 illustrate the results obtained over a 5 year time span, indicating good stability, with a variation in transmittance not exceeding 0.3 percent. Similar results have been observed during the 10 years that such measurements have been made.

Table 3. Stability of Transmittance (T) as a Function of Time, Measured on SRM 930.

Wavelength nm	Percent T			
	1	2	3	Date
440.0	32.87	19.80	11.59	5-18-71
	32.88	19.83	11.62	7-12-72
	32.91	19.81	11.59	8-30-73
	32.96	19.84	11.61	11-16-73
	32.98	19.84	11.62	1- 9-74
	32.94	19.84	11.61	8- 1-74
	32.95	19.84	11.62	1-13-75
465.0	35.53	22.59	13.56	5-18-71
	35.54	22.62	13.59	7-12-72
	35.54	22.62	13.59	8-30-73
	35.62	22.62	13.58	11-16-73
	35.63	22.63	13.58	1- 9-74
	35.60	22.62	13.57	8- 1-74
	35.60	22.62	13.58	1-13-75
590.0	31.13	19.16	10.37	5-18-71
	31.14	19.20	10.41	7-12-72
	31.14	19.20	10.41	8-30-73
	31.21	19.19	10.40	11-16-73
	31.21	19.20	10.41	1- 9-74
	31.18	19.20	10.40	8- 1-74
	31.19	19.20	10.41	1-13-75
635.0	32.55	20.60	11.37	5-18-71
	32.59	20.65	11.39	7-12-72
	32.59	20.65	11.39	8-30-73
	32.66	20.65	11.40	11-16-73
	32.68	20.67	11.41	1- 9-74
	32.63	20.66	11.41	8- 1-74
	32.64	20.66	11.40	1-13-75

Interlaboratory comparisons of the transmittance of SRM 930 were made on the two high-accuracy spectrophotometers at NBS. The results given in Table 4 show that such measurements can be performed within the uncertainty of the instrumental measuring capability. Similarly, measurements made by NBS and by the NPL exhibited a high degree of agreement as illustrated in Table 5.

Table 4. Transmittance of a Set of SRM 930 at Two Wavelengths Obtained Using the Center for Analytical Chemistry (CAC) and the Center for Radiation Research (CRR) High-Accuracy Spectrophotometers. The difference, Δ(CRR), is the difference in transmittance measured by illuminating the filters with polarized radiation having the electric vector in either the horizontal or vertical directions.

Wavelength nm Instrument	Transmittance					
	Nominal 30% Filter		Nominal 20% Filter		Nominal 10% Filter	
	465	546.1	465	546.1	465	546.1
CAC	0.3427	0.3280	0.1980	0.1840	0.1120	0.1014
CRR	0.3430	0.3284	0.1983	0.1844	0.1121	0.1015
Δ(CRR)	-0.00002	-0.00007	-0.00008	-0.00005	-0.00001	-0.00002

Table 5. Comparison Between the Percent Transmittances (%T) Measured on Three Schott NG-4 Glass Filters at NPL and NBS.

Wavelength nm	NBS, %T	NPL, %T	Relative Difference NBS - NPL, %
440.0	12.92	12.93	-0.07
465.0	14.97	15.01	-0.27
590.0	11.67	11.67	0.0
635.0	12.70	12.72	-0.16
440.0	19.60	19.62	-0.10
465.0	22.37	22.43	-0.27
590.0	19.01	19.01	0.0
635.0	20.41	20.47	-0.29
440.0	32.88	32.98	-0.12
465.0	35.53	35.66	-0.36
590.0	31.13	31.21	-0.26
635.0	32.54	32.62	-0.25

Average difference between NBS and NPL percent T values = -0.19 percent.

All the measurements discussed thus far indicate clearly that the glass filters can be used as a secondary transfer optical transmittance standard with an uncertainty of ≤0.5 percent. However, we would like to mention, briefly, a case where the glass material exhibited a greater instability [11]. This incident was recorded on several sets of SRM 930b where an instability with time of about twice the certified uncertainty was observed. This instability was traced to the initial grinding and polishing operation of the glass material by the manufacturer. The problem was eliminated by fine grinding and polishing, and by aging the material in our laboratory for at least six months before its issuance as an SRM. Further information concerning the stability of the optical transmittance of glass filters will be found in a paper by W. R. Blevin [12]. The influence of temperature on the transmittance of glass filters is discussed by A. T. Young [13].

The usefulness of glass filters has been established by the fact that they are used by several manufacturers of spectrophotometers to establish and monitor the accuracy of the transmittance scale of their instruments. To date, about 1000 sets of SRM 930 are in use in this country and abroad.

2.2.3. SRM 931

The limitations of SRM 930 are due to the nature of the glass material which can be used only in the visible spectral range from about 440-700 nm. The need for a material that can be used in the ultraviolet led to the development of the new SRM [14] described below.

SRM 931, illustrated in Figure 12, is a liquid filter consisting of three vials containing a solution of nickel, cobalt, and nitrate ion in dilute perchloric acid and a fourth vial containing only dilute perchloric acid for use as the blank solution. The contents of these vials are transferred to quartz cuvettes with a known radiation pathlength and the transmittances are measured for the three concentrations at 302, 395, 512, and 678 nm using the spectral bandpasses indicated in the certificate. These transmittance values are certified with an uncertainty of 1 percent. SRM 931 has been available from NBS since late 1971, and to date, about 1,050 sets have been sold. Since the use of this liquid SRM requires the availability of cuvettes with a known radiation pathlength, NBS issued a reference cuvette standard (SRM 932) in 1973.

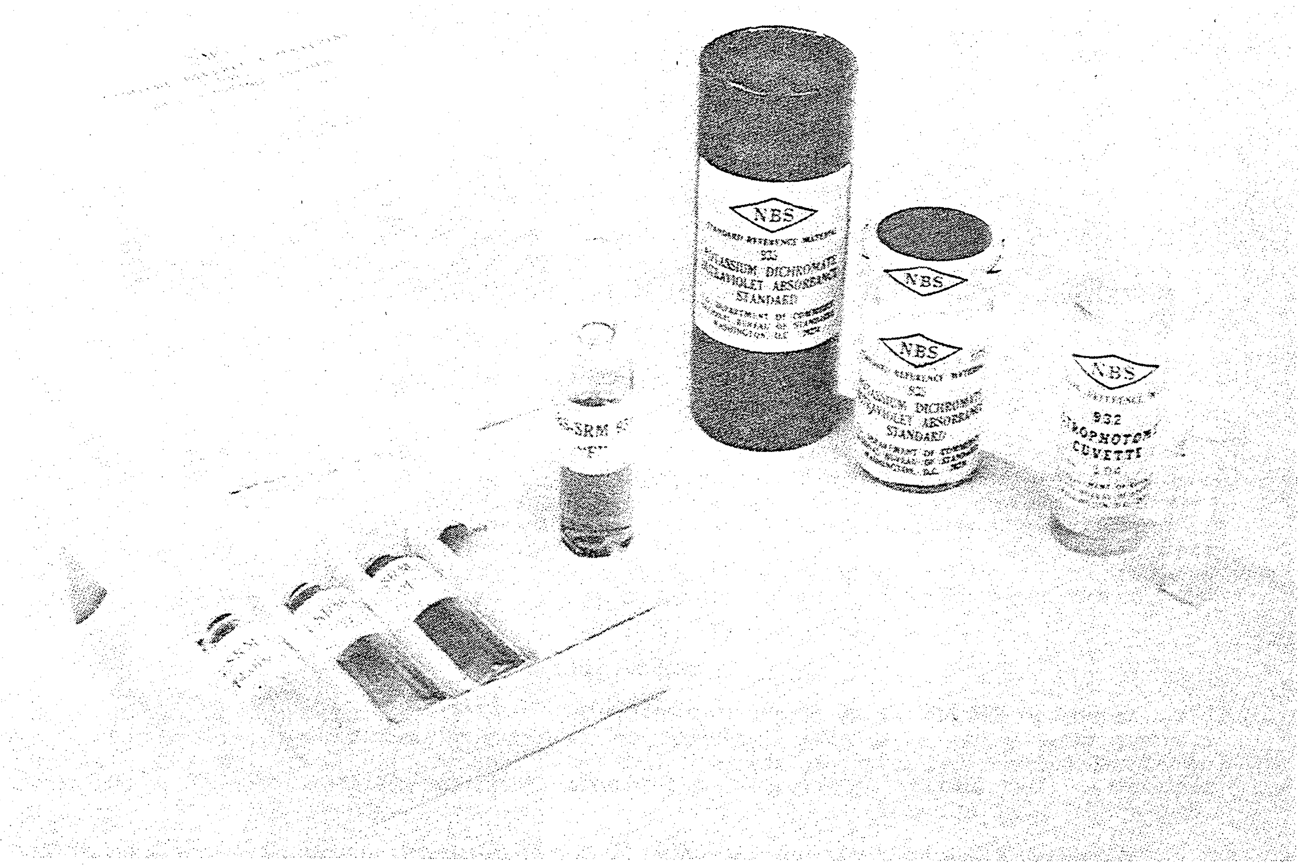

Figure 12. Illustration of SRM 931, Liquid Absorbance Standard; SRM 935, Crystalline Potassium Dichromate Ultraviolet Absorbance Standard; and SRM 932, Quartz Cuvette for Spectrophotometry.

2.2.4. SRM 932

This SRM is illustrated in Figure 13 where it can be seen that the all-fused silica cuvette is made from a rectangular fused silica structure from which segments were cut, ground, and polished as close as possible to 10 mm. Each segment was measured with an uncertainty of 0.5 μm with the instrument shown in Figure 14, and to each face a fused silica window was attached by fusion. Each window is 1.5 mm thick and is flat to 2-3 fringes of mercury green line (546.1 nm) and parallel to 0.002 mm. Each cuvette is provided with a Teflon stopper. The fused silica material is of optical quality and nonfluorescent [15].

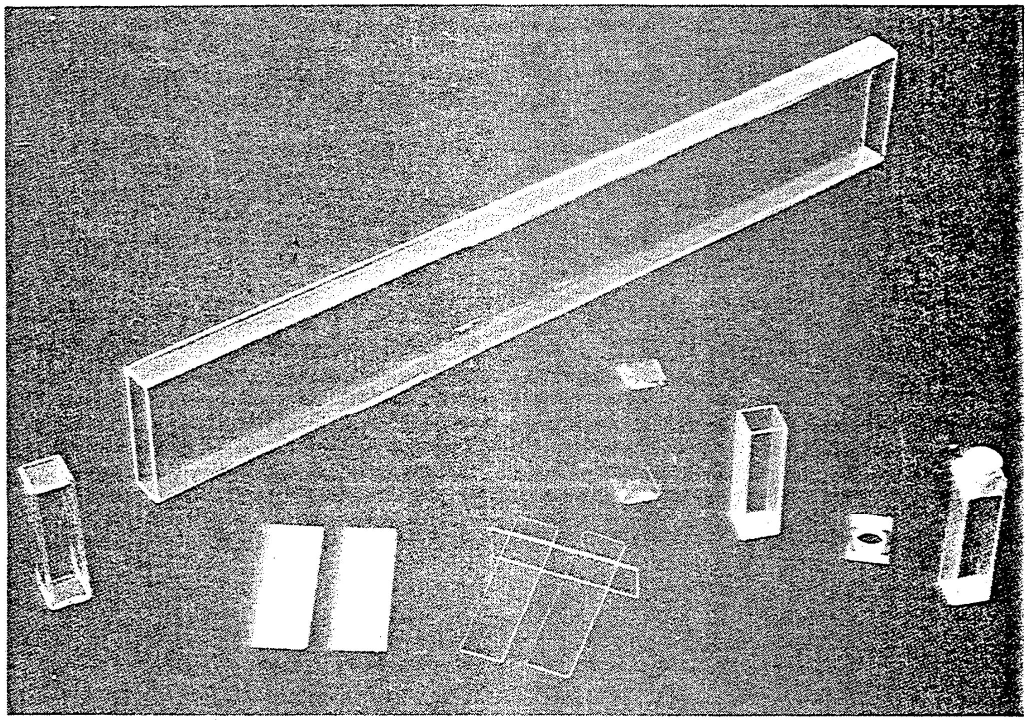

Figure 13. Illustration of SRM 932 Quartz Cuvettes for Spectrophotometry, showing the fused silica material and parts used for constructing the cuvettes for high-accuracy measurements.

The radiation pathlength is measured at 10 positions along the cuvette with a mechanical instrument provided with an electronic lever-type gauge having a resolution of 0.123 µm. The actual usable resolution, however, is 0.312 µm. Figure 15 illustrates this measuring instrument. At the time of calibration, the cuvette pathlengths are certified with an uncertainty of 0.5 µm. The first 100 cuvettes were made in the Glass Shop at NBS. Subsequently, cuvettes were made by a commercial manufacturer following the procedures and specifications established at NBS. All are certified by NBS from measurements made in the Mechanical Processes Division in the Metrology Center.

2.2.5. SRM 935

The use of SRM 931 as an ultraviolet standard is limited to a wavelength of 302 nm. Since numerous measurements below this wavelength are performed in many analytical laboratories, an additional material was selected, studied, and finally issued as an SRM for ultraviolet spectrophotometry. This material is crystalline potassium dichromate dissolved in 0.001 N perchloric acid, and is issued as SRM 935.

This material provides the user with five certified absorbance values in the ultraviolet, and can be used for absorbance linearity verifications at wavelengths near two isosbestic wavelengths, namely 322-323 nm and 345-346 nm. At these points the two chemical species, $HCrO_4^-$ and its dimer $Cr_2O_7^=$, present in the dilute perchloric acid solutions of $K_2Cr_2O_7$, exhibit the same absorptivity.

The wavelength recommended for absorbance linearity measurements is 345 nm where the apparent specific absorbances of the material are sufficiently independent of concentration that absorbance measurements at this wavelength can be used for verifying the absorbance linearity to about one part in a thousand [16].

It is appropriate to remind spectrophotometrists that absorbance linearity should not be equated with transmittance accuracy. Indeed, as discussed by F. J. J. Clarke "the absorbance linearity or obedience to Beer's law, establishes only that the photoelectronic system obeys a simple power law, not that the power (exponent) is exactly unity" [5]. It must also be kept in mind that, theoretically, Beer's law is valid only at infinite dilutions.

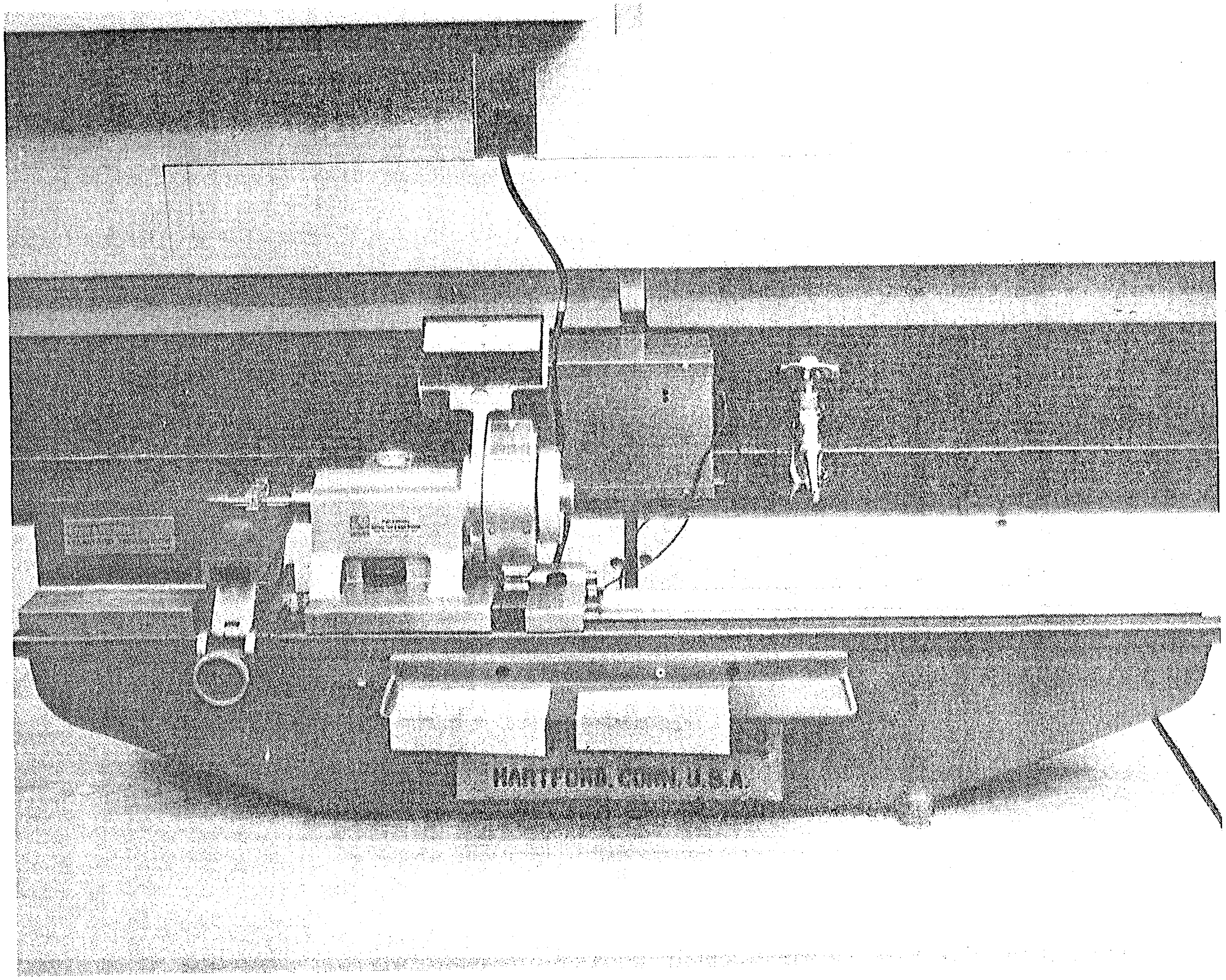

Figure 14. Instrument for measuring the thickness of window plates and length of fused silica segments.

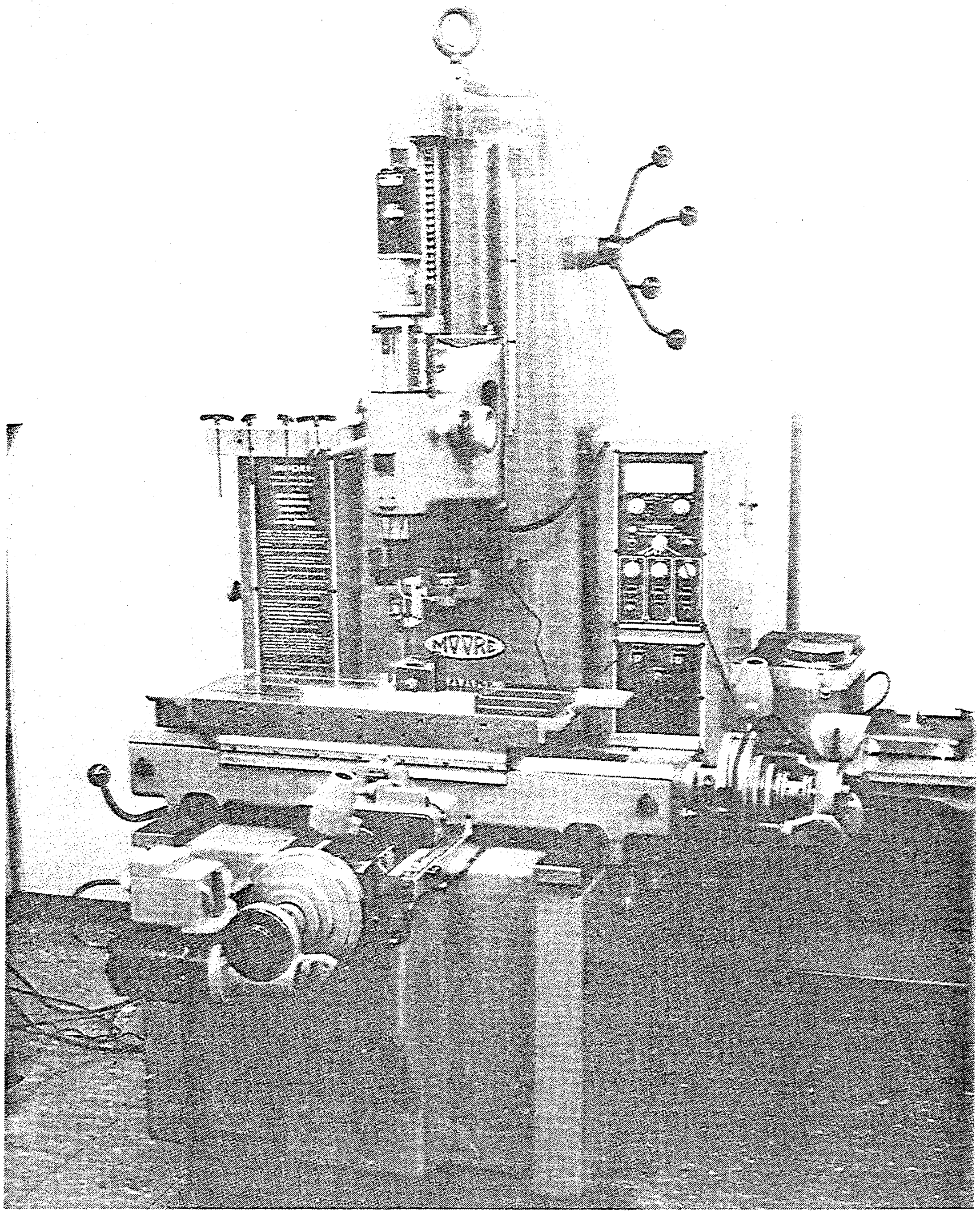

Figure 15. General view of the standard instrument used to measure the radiation pathlength of assembled cuvettes.

The crystalline potassium dichromate, of known purity and homogeneity, is issued in a nonactinic brown glass bottle where it should be kept at room temperature at all times. This SRM is illustrated in Figure 12. In certifying this material, solutions were prepared in 0.001 N perchloric acid and certified for their apparent specific absorbance at five concentrations between 0.020 to 0.100 g · kg^{-1} and at wavelengths of 235, 257, 313, 345, and 350 nm. Fused silica cuvettes with a nominal radiation pathlength of 10 mm and known to 0.5 μm were used in all measurements. The term "apparent" is used throughout both the certificate and the NBS SP260-54 publication to indicate that the values are not corrected for the effect of internal multiple reflections within the cuvette, nor have the weights been corrected to vacuum. The accurate specific absorbances of acidic potassium dichromate solutions, corrected for these two parameters are given in Reference 16. Since the radiation pathlength of the cuvettes is a critical factor in the measurements, it is recommended that the NBS certified fused silica cuvettes SRM 932 be used with this SRM 935. The uncertainty in the apparent specific absorbance ranged from 0.034 g · kg^{-1} at the lowest concentration to 0.010 g · kg^{-1} at the higher concentration.

2.2.6. Limitations of SRM's 930, 931, and 935

While each of the SRM's described thus far are useful and have positive qualities, they suffer from certain inherent limitations. The glass filters, SRM 930, can be used only in the visible spectral range; liquid standard SRM 931 provides only one calibration point in the ultraviolet; and the potassium dichromate, SRM 935, requires that accurate solutions be prepared, and that cuvettes with accurate radiation pathlength be available. Furthermore, this material is useful for providing calibration data only in the ultraviolet. In addition, a common limitation of these SRM's, is that they require the use of spectrophotometers capable of providing narrow spectral bandpasses.

2.2.7. SRM 2031

To eliminate these limitations, a number of materials were studied to determine their usefulness as transmittance SRM's that could be used in the ultraviolet and visible spectral regions with little dependence on spectral bandpass, and could be used directly without any laboratory preparation. The material finally selected was the semitransparent evaporated metal-on-fused silica filters. Although this material was available from the early stage of the project, the problem of protecting the exposed surface of the metal layer from deleterious environmental action had to be solved before further work could be undertaken to determine the suitability of this material as a transmittance SRM [17]. The protection of the exposed metal semitransparent layer was achieved by covering the surface with a clear fused silica plate applied by optical contact. Under these circumstances the fused silica plate carrying the evaporated metal layer with the clear plate behave optically as if they were a single solid semitransparent block. A detailed study was undertaken to assess the optical transmittance properties such as: spectral transmittance, stability, scatter, polarization, uniformity, position dependence, aging, capability of interlaboratory exchange of measurements. Based on the results obtained in this study, the material was issued as an SRM for ultraviolet and visible spectrophotometry, and is illustrated in Figure 16.

The standard consists of three filters, all made from nonfluorescent, optical grade fused silica material. Two of the filters are made from a sandwich of two fused silica plates, each 1.5 mm thick, one plate carrying the semitransparent evaporated metal layer, while the other plate is transparent. The plates are ground and polished to achieve optical contact. The third filter is made from a single plate of transparent material 3 mm thick. The filters have a nominal transmittance of 10, 30, and 90 percent. They are mounted in metal holders provided with plastic shutters for mechanical protection. A metal cylindrical box is used to store the filters, and an additional empty filter holder is provided to be used in the reference beam of the spectrophotometer, to insure that identical measurement conditions are achieved.

This SRM is usually certified at the following 10 wavelengths: 250, 280, 340, 360, 400, 465, 500, 546.1, 590, and 635 nm. Transmissions at additional or different wavelengths can be provided on request. The uncertainty of the certified transmittance is 1.0 percent.

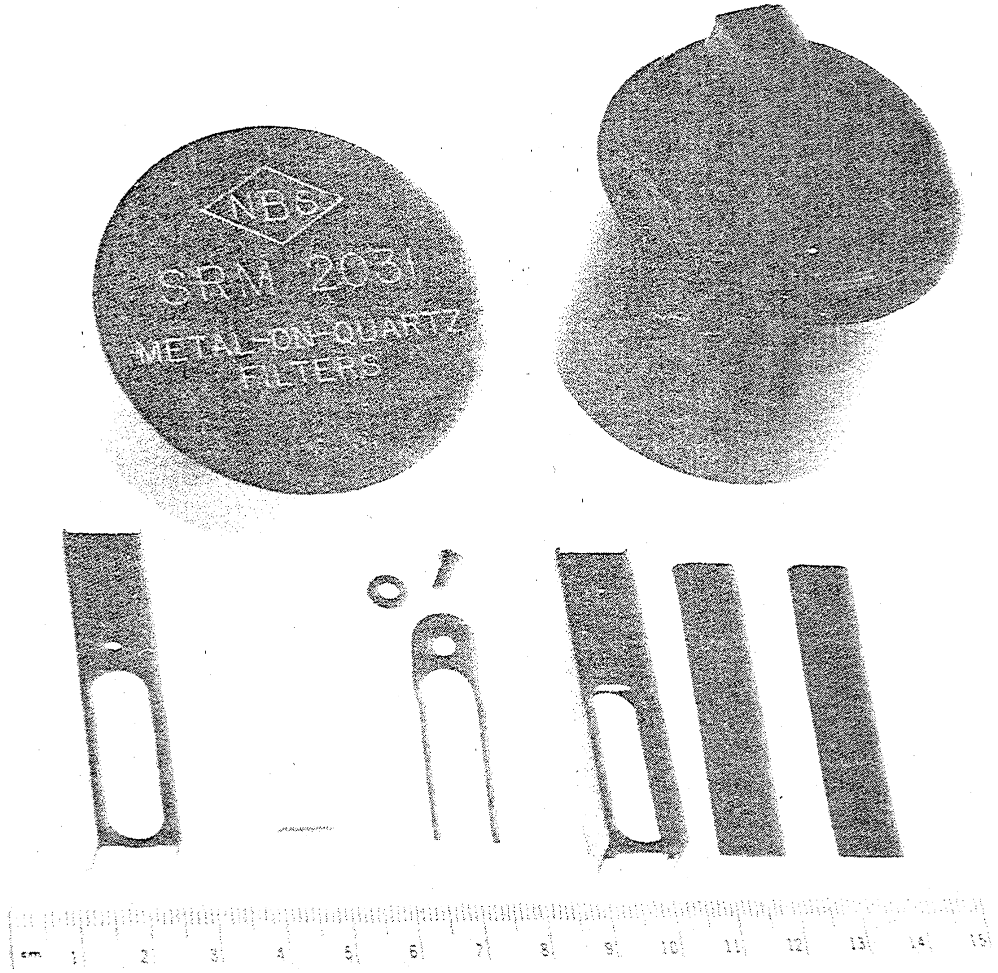

Figure 16. Illustration of SRM 2031, Metal-on-Quartz Filters for Spectrophotometry. Top: Cylindrical container with screw cap, both made of black anodized aluminum alloy. Four filter holders can be stored in the container. Bottom: (from left to right): Aluminum alloy filter holder; metal-on-fused silica filter; retaining spring with nylon screw and washer; assembled unit and two plastic shutters. All metal and plastic parts are flat black.

Of the numerous tests performed on this SRM, only the measurements performed by K. Eckerle at NBS using the high-accuracy spectrophotometer in the Center for Radiation Research will be described. From the data in Tables 6 and 7 it can be concluded that, within the experimental measuring conditions, there is no significant change of transmittance due to scatter. The material is homogenous and no polarization effect could be detected. Results of interlaboratory measurements on the same filters by NBS and the Naval Weapons Center are illustrated in Table 8 which shows that the comparative transmittance could be achieved with an uncertainty similar to that of the performance of the instruments themselves.

Table 6. Transmittance of Some Inconel-on-Fused Silica Filters Obtained Using the Center for Analytical Chemistry (CAC) and the Center for Radiation Research (CRR) High-Accuracy Spectrophotometers. The difference, Δ(CRR), is the difference in transmittance measured by illuminating the filters with polarized radiation having the electric vector in either the horizontal or vertical directions.

	Transmittance									
Wavelength nm / Instrument	250	300	340	400	440	465	500	546.1	590	635
					Nominal 30% Filter					
CAC 9/75	0.2867	0.2989	0.3017	0.2992	0.2963	0.2949	0.2938	0.2936	0.2940	0.2948
CRR 8/76	.2879	.2997	.3025	.3001	.2974	.2960	.2948	.2946	.2950	.2958
CAC 10/76	.2880	.2997	.3026	.3000	.2973	.2958	.2947	.2946	.2950	.2958
Δ(CRR)	0.00013	-0.00002	0.00002	0.00000	0.00005	0.00004	0.00001	0.00010	0.00008	0.00002
					Nominal 20% Filter					
CAC 9/75	0.2127	0.2115	0.2023	0.1884	0.1824	0.1800	0.1781	0.1774	0.1778	0.1787
CRR 8/76	.2129	.2117	.2023	.1885	.1825	.1802	.1782	.1776	.1779	.1788
CAC 10/76	.2131	.2118	.2023	.1885	.1825	.1802	.1784	.1778	.1781	.1790
Δ(CRR)	0.00004	0.00011	0.00002	0.00001	0.00008	0.00002	0.00004	0.00005	0.00007	-0.00002
					Nominal 90% Filter					
CAC 9/75	0.9127	0.9231	0.9265	0.9291	0.9300	0.9305	0.9312	0.9315	0.9320	0.9324
CRR 8/76	.9124	.9225	.9263	.9293	.9303	.9307	.9312	.9318	.9322	.9325
CAC 10/76	.9124	.9224	.9262	.9287	.9298	.9304	.9308	.9314	.9320	.9324
Δ(CRR)	-0.00016	0.00014	0.00002	-0.00015	0.00006	0.00009	0.00014	0.00011	0.00001	-0.00028

Table 7. Measurement of Scatter, Homogeneity, and Polarization on Three Inconel-on-Fused-Silica Filters[a].

(x, y)	Ω	T(0°)	$\Delta T_R(0°)$	T(90°)	$\Delta T_R(90°)$	T(AVE)	ΔT_R(AVE)
			#20 Filter 1				
(+1,0)	0.0045	0.10807	0.000018	--	--	--	--
(-1,0)	0.0045	0.10798	0.000009	--	--	--	--
(0,0)	0.0045	0.10805	0.000013	0.10841	0.000020	0.10823	0.000012
(0,0)	0.0012	0.10801	0.000027	0.10840	0.000020	0.10821	0.000017
(0,0)	0.014	0.10799	0.000010	0.10831	0.000013	0.10815	0.000008
			#20 Filter 2				
(+1,0)	0.0045	0.32089	0.000035	--	--	--	--
(-1,0)	0.0045	0.32050	0.000037	--	--	--	--
(0,0)	0.0045	0.32066	0.000046	0.32092	0.000030	0.32081	0.000027
(0,0)	0.0012	0.32069	0.000039	0.32079	0.000045	0.32073	0.000030
(0,0)	0.0014	0.32045	0.000047	0.32050	0.000057	0.32047	0.000037
			#20 Filter 3				
(+1,0)	0.0045	0.93139	0.000118	--	--	--	--
(-1,0)	0.0045	0.93160	0.000096	--	--	--	--
(0,0)	0.0045	0.93131	0.000067	0.93125	0.000064	0.93128	0.000046
(0,0)	0.0012	0.93107	0.000122	0.93095	0.000039	0.93101	0.000064
(0,0)	0.014	0.93089	0.000041	0.93076	0.000080	0.93083	0.000045

[a] NOTE: All measurements were performed at λ546.1 nm. The quantities (x,y) are the coordinates of the center of the illuminating spot with respect to the center of the filter in millimeters when looking in the direction of the source. The parameter, Ω, is the approximate solid angle in steradians. The quantities T(0°), T(90°), and T(AVE) are the transmittances for the electric vector of the illuminating radiation horizontal, vertical, and average of horizontal and vertical, respectively, measured at λ546.1 nm. Their respective standard errors are $\Delta T_R(0°)$, $\Delta T_R(90°)$, and ΔT_R(AVE). The radiation scatter measurements were made by determining the transmittance of the filters successively at distances from the averaging sphere of 210 mm, 370 mm, and 715 mm corresponding to angles of about 0.014, 0.0045, and 0.0012 steradians, respectively. The diameter of the circular entrance aperture at the averaging sphere was 28 mm. The transmittance measurements to assess the uniformity of the filters were made at a constant angle of 0.0045 steradians.

Table 8. Comparative Measurements of Transmittance on Inconel-on-Fused Silica Filters in the Ultraviolet and Visible from 250 nm to 635 nm at NBS and at Naval Weapons Center (NWC).

		Transmittance, Percent									
Filter No.	Lab.	250	300	340	400	440	465	500	546.1	590	635
45	NBS	1.492	1.844	1.899	1.973	2.083	2.171	2.318	2.540	2.750	2.956
	NWC	1.501	1.837	1.884	1.953	2.070	2.158	2.317	2.552	2.755	2.958
46	NBS	21.49	21.39	20.46	19.05	18.45	18.20	18.01	17.94	17.98	18.07
	NWC	21.50	21.37	20.39	19.04	18.38	18.19	18.06	17.99	17.92[a]	18.10
47	NBS	28.73	29.96	30.24	29.99	29.70	29.56	29.44	29.41	29.45	29.55
	NWC	28.80	29.89	30.23	29.95	29.68	29.55	29.45	29.42	29.43	29.48
48	NBS	91.25	92.28	92.64	92.91	93.00	93.04	93.10	93.15	93.19	93.22
	NWC	91.21	92.22	92.59	92.90	92.98	93.01	93.14	93.19	93.21	93.24

[a] Value corrected as indicated in J. M. Bennett communication.

2.2.8. SRM's 2009, 2010, 2013, and 2014

The SRM's discussed thus far are intended to verify the accuracy of transmittance measurements and the radiation lightpath. Certified materials to verify the wavelength accuracy and to assess the stray radiant energy are also available from NBS.

The SRM's for verifying the wavelength accuracy of conventional spectrophotometers are SRM's 2009, 2010, 2013, and 2014. They were calibrated by Venable and Eckerle of the NBS Center for Radiation Research [18]. Each of these standards consist of a didymium glass filter having a nominal spectral transmittance shown in Figure 17. These filters are produced in two nominal sizes: 51 mm x 51 mm x 3 mm thick and 10 mm x 30 mm x 3 mm thick. They are mounted in a metal holder that fits into the cuvette holder of the spectrophotometer. They are individually calibrated (SRM's 2013 and 2014) or batch calibrated (SRM's 2009 and 2010), and, depending on the accuracy desired, one or the other SRM should be used. The spectral bandpass-wavelength dependence is described in detail in the certificate that accompanies each standard. The 14 to 24 certified wavelengths are in the spectral range 400-760 nm and are given for spectral bandpasses from 1.5-10.5 nm.

2.2.9. SRM's 2032 and 2033

An instrumental parameter that can measurably affect the accuracy of transmittance is the stray radiant energy (SRE) or stray light. This can be defined generally as unwanted radiation that is measured by the photodetector, and contributes to the degradation of optical transmittance measurements, and to the reduction of specificity, sensitivity, and linearity of absorbance measurements. Two major types of SRE can be identified: heterochromatic and isochromatic. The former originates in the spectrophotometer and can be defined as radiation transmitted by the spectrometer that is of a wavelength different from the spectral bandpass for which the instrument was set. The latter originates in the sample compartment and can be defined as radiation of the same wavelength as that which falls on the sample, but reaches the photodetector of the spectrophotometer without passing through the analytical sample.

From these simple definitions it might be concluded that both stray lights can be assessed easily. Unfortunately, this is not the case. Actually, the accurate measurement of stray light is subject to ambiguity and is difficult to define and measure. This situation is discussed in some detail in an NBS Special Publication, "Stray Radiant Energy in Ultraviolet Molecular Absorption Spectrophotometry, SRM's 2032 and 2033", R. Mavrodineanu and R. W. Burke (in press).

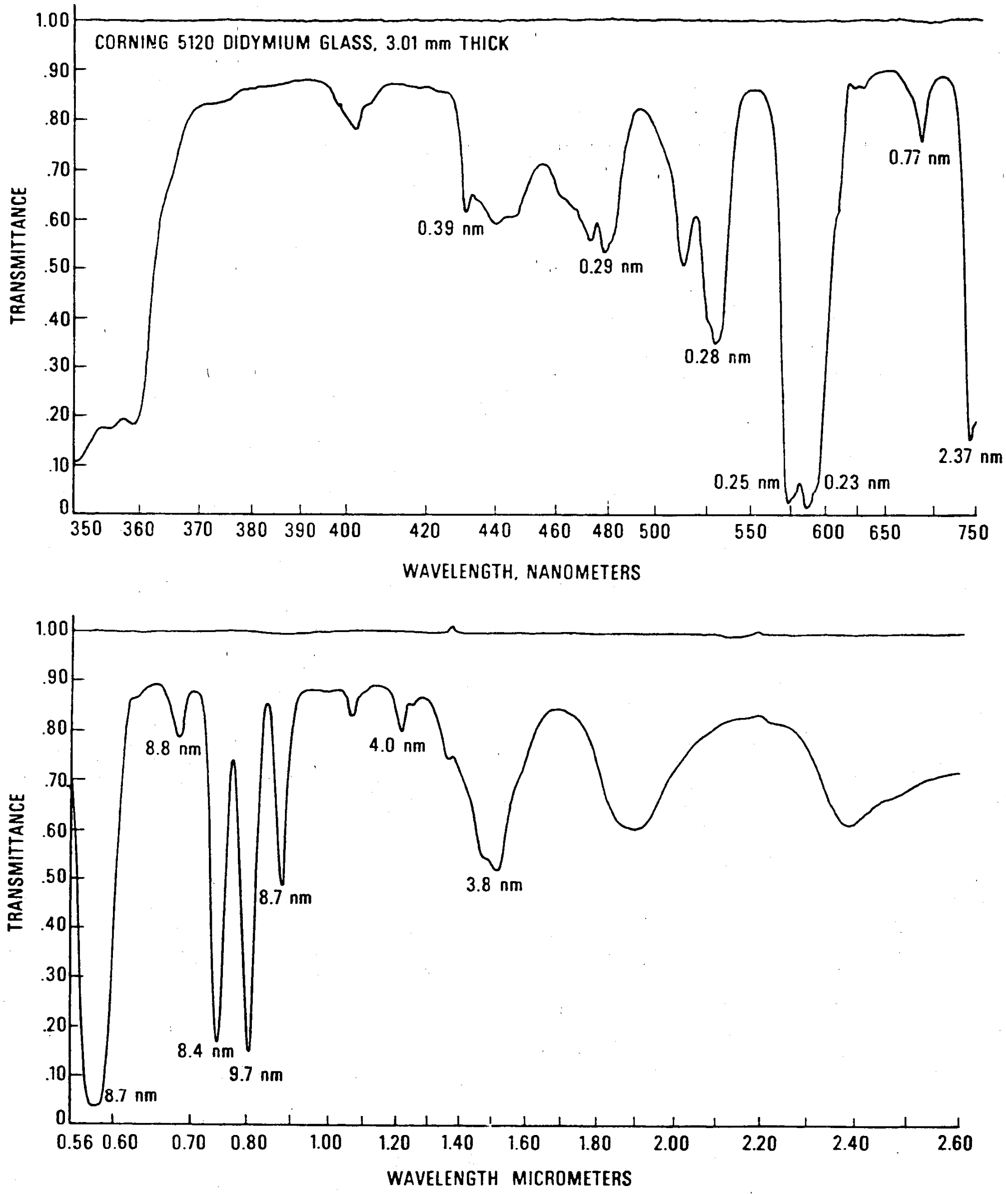

Figure 17. Transmittance of a didymium glass filter recorded at the Corning Glass Works. The spectral bandpass used is marked at the corresponding transmittance minimum.

At this time, two SRM's (SRM's 2032 and 2033) are available to assess both the heterochromatic and isochromatic stray light. Heterochromatic stray light can be measured by using a variety of techniques, including the blocking optical filters, and this approach was selected to produce SRM 2032. This material consists of crystalline potassium iodide of reagent-grade purity which has sharp cutoff absorption near 260 nm, and can be used to assess the stray light below this wavelength. Solutions from this compound were certified for their specific absorbances at eight wavelengths from 240-275 nm in increments of 5 nm with an uncertainty of 5 percent. The measurement consists of setting the spectrometer at a wavelength below 260 nm, say 255 nm, and any appreciable amount of light detected is heterochromatic stray light.

Isochromatic stray light can be assessed with the help of SRM 2033. This material consists of two filters made from a semitransparent evaporated metal on a fused silica substrate each of which has a nominal transmittance of 10 percent. One filter is mounted in a metal holder that can be inserted in the sample compartment of the spectrophotometer. This holder is provided with a front and rear shutter, one of which has a window in which the second filter is mounted, as illustrated in Figure 18. This system is used to assess the maximum isochromatic stray light in the sample compartment that results from reflections of the incident radiation at the surfaces of the sample and various optical components and reaches the photodetector without passing through the sample.

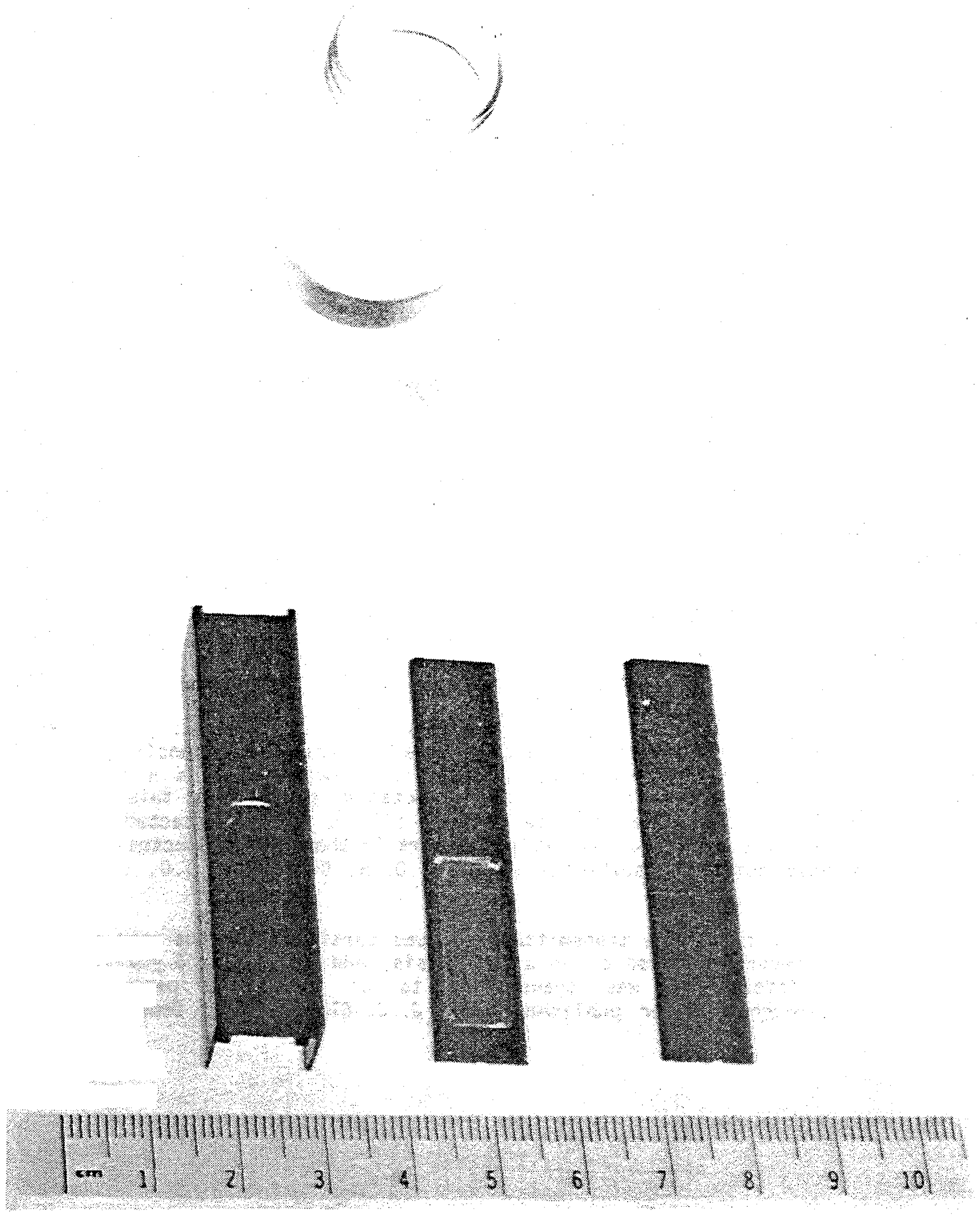

Figure 18. Illustration of the radiation attenuator consisting of a metal filter holder provided with a filter made of a semitransparent evaporated metal-on-fused silica plate. Two plastic shutters are also shown, one is provided with a second filter consisting of a semitransparent evaporated metal-on-fused silica plate. Each filter has a nominal transmittance of 10 percent. The other shutter is opaque. The unit is stored in the plastic container shown in the figure.

The measurement is performed by placing this unit in the sample beam of the spectrophotometer with the opaque shutter at the rear of the filter holder. Under these conditions, if a signal is detected, it is caused by reflections at the surface of the filter exposed to the incident radiation. This radiation is scattered from the walls and other components of the sample compartment and reaches the photodetector without passing through the sample. This signal is the isochromatic stray light.

Before concluding this summary description of the SRM's developed at NBS in the field of spectrophotometry, we would like to mention the availability of an SRM for fluorescence measurements. This material, SRM 936, quinine sulfate dihydrate is certified for the relative molecular emission spectrum in radiometric units from 375-675 nm at 5 nm intervals. The certification was made by R. A. Velapoldi and K. D. Mielenz using a specially developed reference fluorescence spectrometer built by K. D. Mielenz [19,20].

3. Future Developments in the Field of SRM's for Spectrophotometry

The program at NBS for the continuation of basic research in the field of high-accuracy spectrophotometry and for production of other SRM's in this field is active, and the following projects are presently being considered:

- Production of a wavelength standard having reference values in the ultraviolet and having sharper absorption peaks, such as those exhibited by holmium oxide solutions in perchloric acid.
- Basic study on the nature and measurements of stray radiant energy, and development of additional SRM's in this field, extending the assessment of this parameter closer to the visible, i.e., 400 nm.
- Extending the certification of solid or liquid transmittance standards into the near-infrared spectral region. This will require the extension of the measuring capabilities of the NBS high-accuracy spectrophotometers into the near-infrared to 3 µm.

The implementation of these and other projects in this field depends on the availability of resources and the demand from the users of spectrophotometers.

4. Transmittance Transfer Standards from the National Physical Laboratory in Great Britain

The National Physical Laboratory in Teddington, Great Britain, is issuing on a regular basis semitransparent evaporated metal filters on fused silica substrates as transfer standards in the ultraviolet, visible, and near-infrared spectral range. The metal used is a specially selected nichrome alloy. These filters are certified at any selected densities over this spectral range as required by the user. The exposed metal surface of these filters is not protected. Neutral optical glasses of the Schott NG type are also available for work in the visible spectral range, and those available from stock have nominal absorbances of 0.04, 0.15, 0.25, 0.5, 1.0, 1.5, 2.0, 2.5, and 3.0.

To preserve the integrity of the transmittance values certified for the metal-on-fused silica filters, NPL recommends recertification on an annual basis, and every 2 or 3 years for the neutral glass filters. This information was transmitted to us through a personal letter from F. J. J. Clarke, and through a paper published by F. J. J. Clarke, M. J. Downs, and W. McGivern [21].

PART TWO

5. Spectrometric Applications

Ultraviolet and visible spectrophotometry continues to be used extensively for the chemical characterization of numerous types of materials. Today, as in the past, practically all of the measurements used for chemical analyses are made on liquid samples. for these measurements to be meaningful and the compositions subsequently calculated to be accurate, proper attention must be given in general to the operational aspects of the technique and, specifically, to the selectivity of the method used. Recommendations involving the cuvette and the importance of maintaining good temperature control of the sample during measurement are presented in the two sections that immediately follow. These discussions are followed by concise descriptions of several spectrophotometric methods that have been in use at NBS for a number of years and are cited here because of their high selectivity.

6. Experimental

6.1. Cuvettes

The proper selection, care, use, and manipulation of cuvettes is one of the most overlooked operations in the spectrophotometric measurement process. Improper handling of the sample cuvette has, in many instances, undoubtedly contributed to the uncertainty of the results. A recent paper by Rothman et al., [22] has shown that the imprecision of cuvette positioning can be the limiting factor in making precise measurements.

The manufacturers have done a commendable job in making available quality cuvettes at a modest cost. It is the users' responsibility to avoid any operations that might degrade this quality. The rectangular, nonfluorescent, fused quartz cuvette with fused edges and a tightly-fitting circular stopper, preferably made from tetrafluoroethylene polymer, is the most desirable and useful type. Upon purchase of a new cuvette, the user should immediately engrave a permanent mark on its side near the top so that measurements are always made with the same orientation of the cuvette in its holder.

Numerous procedures have been used for cleaning cuvettes. These include dilute hydrochloric acid, mild detergent in water, water and alcohol, ethanol-nitric acid, sulfuric acid, and sulfuric acid containing potassium dichromate or potassium permanganate. For most applications, our experience has been that the use of distilled water and ethanol is generally adequate, especially if the cuvette is emptied and cleaned immediately after use. If a stronger cleaning measure is required, immersing the cuvette in either concentrated or, on occasion, fuming sulfuric acid is preferred. Recently, Goddard [23] has recommended a hot solution of trisodium orthophosphate as providing the most satisfactory and practical cleaning of cuvettes for use in the ultraviolet (see also reference 10).

As regards cuvette positioning, highest precision is obtained when the cuvette is placed in the sample holder within the sample compartment of the spectrophotometer and left in that holder until a set of absorbance measurements on a series of standards and sample solutions has been completed. For transferring solutions to and from the cuvette, the Pasteur-type, disposable pipettes have been found to be most useful. Both soft and borosilicate glass pipettes are widely available. In general, the soft glass type should not be used since their surface is sufficiently alkaline to affect the absorbance of pH-sensitive solutions [24]. All measurements are made relative to air and the absorbance of the sample is determined by subtracting the apparent absorbance of the blank from the solution value. The data in Table 9 illustrate the typical sequence of measurements and the level of precision obtainable on the NBS high-accuracy spectrophotometer. The measurements were made on a set of acidic potassium dichromate solutions used in the certification of the crystalline salt as an SRM [16].

Table 9. Reproducibility of Absorbance Measurements at 235 nm for a Cycle of Solvent and Sample Runs.

Cuvette No.	Solvent, after 5 rinses	Sample, after 5 rinses	Sample, after 7 rinses	Solvent, after 5 rinses
14	0.05198	0.30128	0.30141	0.05213
16	0.05099	0.55052	0.55057	0.05093
18	0.05310	0.80066	0.80074	0.05321
25	0.05002	1.04863	1.04855	0.05022
26	0.05064	1.30351	1.30333	0.05065
Reference filter[a]	0.55913	0.55925	0.55936	0.55922

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[a] Inconel on quartz.

The agreement between absorbance measurements made on a set of acidic potassium dichromate solutions in October 1975 and again in June 1977 (Table 10) illustrates, rather dramatically, the quality of results that can be obtained with accurate instrumentation, stable samples, calibrated cuvettes, and good technique.

Table 10. Absorbances of a Set of 0.001 N Perchloric Acid Solutions of SRM 935 $K_2Cr_2O_7$ at Time of Preparation and After 20 Months Storage.

Nominal $K_2Cr_2O_7$ Concentration	257 nm (max)		350 nm (max)	
	1975	1977	1975	1977
20	0.2872	0.2871	0.2145	0.2150
40	0.5773	0.5760	0.4305	0.4307
60	0.8674	0.8677	0.6456	0.6450
80	1.1623	1.1608	0.8612	0.8616
100	1.4559	1.4565	1.0773	1.0755

6.2. Temperature Control

The importance of controlling temperature, particularly during the spectrophotometric analysis of liquid samples, is well established. Temperature coefficients of absorbance of 0.1 to 0.2 percent per degree Celsius are not unusual. As a rule, at least 2 minutes equilibration time should be allowed for every degree Celsius above or below ambient that the measurement is performed. In addition, if measurements are made more than ±5 °C from ambient, the analyst must be aware of potential differences in bath and sample compartment temperatures, even at equilibrium, because of losses from the supply lines. Some data for a typical thermostating assembly are given in Table 11. The temperatures were measured by placing one end of an iron-constantan thermocouple in the cuvette solution and the other end in the constant temperature bath.

Table 11. Temperature Differences Between Thermostating Bath and Cuvette Solution; Ambient T = 24.2 °C.

Bath Temperature, °C	Cuvette Solution, °C	Temperature Differences, °C
15.0	17.4	+2.4
20.0	21.4	+1.4
25.0	24.9	-0.1
30.0	29.1	-0.9
35.0	32.9	-2.1
40.0	37.0	-3.0

7. Selected Methodologies

This section describes some of our experiences in applying spectrophotometric measurements to materials characterization, particularly for the trace inorganic characterization of NBS SRM's. A comparison of the spectrophotometric values with those obtained by isotope dilution mass spectrometry (IDMS), as well as by other trace instrumental techniques, has been used to estimate the bias of the spectrophotometric data.

7.1. Antimony and Arsenic

The spectrophotometric determination of antimony with brilliant green (BG) and of arsenic with silver diethyldithiocarbamate (AgDEDC) are probably the best wet chemical techniques for the determination of trace amounts of these elements. These methods, as employed for the analysis of both iron and copper-base alloys, require that the sample be dissolved under conditions such that antimony and arsenic are obtained in their +3 oxidation states. This is achieved by dissolving the iron or copper alloy in concentrated hydrochloric acid in sealed borosilicate ampoules at a temperature of 200 to 250 °C. The principal steps in the two procedures are summarized in Figure 19.

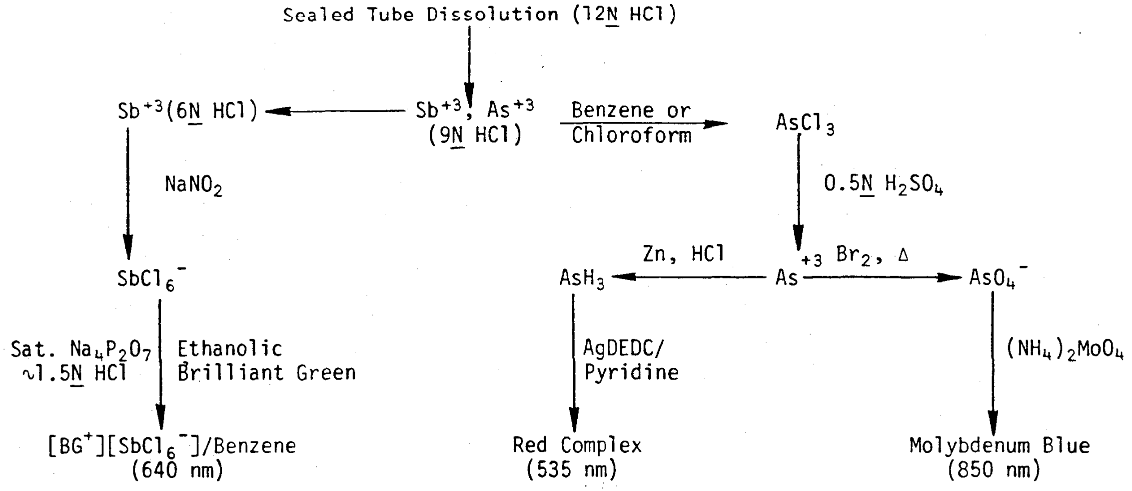

Figure 19. Outline of the spectrophotometric procedures used for determining antimony and arsenic in NBS iron and copper base alloys.

The need for certifying antimony in iron base NBS SRM's was initiated by the general opinion among metallurgists in the early 1960's that trace amounts of this element were responsible for the cracking failures of rotor steels. Although this theory for the most part was subsequently disproven, the original procedure [25] has been used unchanged except for one modification namely preparing the stock solution of brilliant green in ethanol rather than in water. Two important advantages were gained by this change. In the first place, the stability of the dye solution was increased from one or two days to several months. Secondly, the use of ethanol eliminated the hypsochromic shift and decreased intensity of the absorbance maximum that had been observed previously when sodium nitrite was used as the oxidant (Figure 20). Nitrite was, and continues to be, the preferred oxidant because close control of its concentration is not as critical as it is for cerium (IV) or hydrogen peroxide. Experiments performed to understand the role of ethanol indicate that, in its absence, nitrite interacts with the dye, most probably by addition of a nitro group to one or more of the ring systems. If ethanol is present, however, when the dye is added, or if the dye is added as an ethanolic solution, then the excess nitrite remaining after oxidation of Sb(III) to Sb(V) is apparently converted into a nonreactive alkyl derivative (CH_3CH_2ONO).

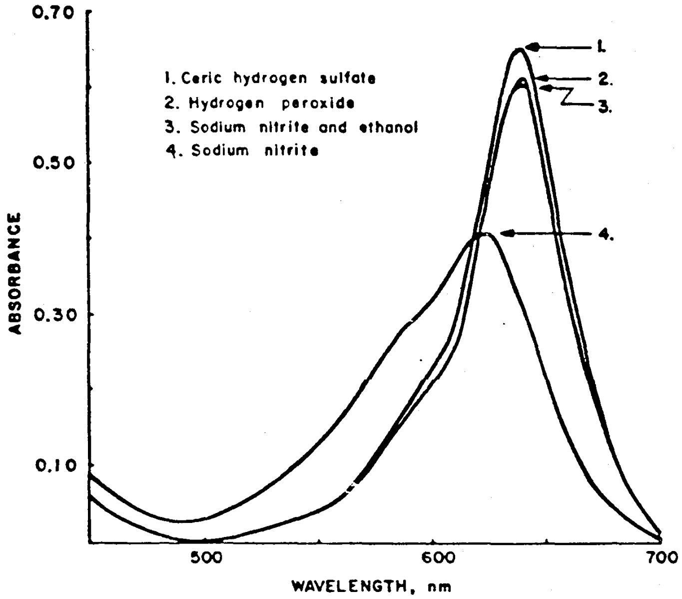

Figure 20. The effect of various oxidation procedures on the absorbance spectra of the brilliant green-hexachloroantimonate complex.

The spectrophotometric determination of arsenic based on the color reaction between arsine and silver diethyldithiocarbamate is presently being used extensively in the analysis and certification of NBS biological and botanical SRM's. Although the exact composition of the colored product is not known, the method is straightforward and simple to run. The minimum amount of arsenic that can be determined is about 0.05 µg. Stibine and phosphine interfere by also giving colored products with AgDEDC. However, their spectra are sufficiently different that their presence may be checked by making absorbance measurements at two wavelengths. If only arsenic is present, the ratio of the absorbance at 505 nm to that obtained at 535 nm (λ_{max} for arsenic complex) should not exceed 0.82. The decision to use the arsine-AgDEDC procedure or the molybdenum blue method (Figure 19) is dictated by the amount of arsenic present. For more than 10 µg, the latter method is preferred.

In a subsequent publication [26], the possibility of coupling the selective extraction of arsenic (III) with the arsine-AgDEDC method for doing speciation measurements of As(III) and As(V) in organic materials was explored. Although the method works well for aqueous mixtures and may be useful for speciation measurements on natural water samples, it is presently not applicable to the analysis of organic materials until a digestion medium is found in which the original valence states of arsenic are not altered. Using several mild, nonoxidizing acid media, some As(V) was invariably reduced to As(III), apparently by other solubilized constituents in the samples.

An important application of the antimony and arsenic procedures has been the determination of these elements in the NBS copper benchmark SRM's. The issuance of this series of unalloyed copper standards will be the culmination of one of the largest undertakings of the NBS Standard Reference Materials Program. More than 70 participants cooperated in the planning of these SRM's, 16 companies contributed base materials, and 22 laboratories have performed cooperative analyses. Currently 11 copper SRM's containing varying levels of trace impurities are available from the NBS Office of Standard Reference Materials.

The NBS values for antimony and arsenic in SRM's 394, 395, and 396 are listed in Table 12 together with the average values obtained from cooperating laboratories.

Table 12. Antimony and Arsenic in NBS Copper Benchmark SRM's.

	Values by Weight, ppm			
	NBS Values		Average of Cooperating Labs.	
SRM No.	Sb	As	Sb	As
394 (Cu I)	4.8[a]	2.6[a]	5.6	3.0
395 (Cu II)	7.9 (8.3)[b]	1.8 (2.4)[b]	7.0	2.2
396 (Cu III)	<0.5 (0.16)	<0.5 (≤0.03)	2.0	1.5

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[a] Spectrophotometric values.

[b] Values in parentheses were obtained by spark source mass spectrometry.

The spark source results were calculated on the assumption that the spectrophotometric values on 394 are accurate. Paulsen [27] has since reported a value of 4.5 ppm for Sb in 394 by isotope dilution-spark source mass spectrometry. Antimony has not been determined to date in 395 or 396 by isotope dilution and arsenic cannot be determined by isotope dilution because it is monoatomic.

The data reported by the cooperating laboratories are based on optical emission methods. Their average values are in good agreement with the NBS results for 394 and 395. However, for 396, their average value for both antimony and arsenic appear to be at least an order of magnitude high. This discrepancy may be caused by the optical emission methods being used at or near their detection limits or the standards used containing less antimony and arsenic than assumed.

7.2. Nickel

The spectrophotometric determination of nickel by the persulfate-dimethylglyoxime procedure has been studied extensively by Deardorff and Menis [28] and they have applied it exhaustively to the analysis of a wide variety of NBS SRM's. Although the exact nature of the colored complex is not known, present evidence suggests that nickel is oxidized to the +4 oxidation state. The red complex forms almost instantaneously when ammonium persulfate is added to a nickel solution containing citrate ion, the pH is adjusted to 13, and alcoholic dimethylglyoxime is added. The presence of citrate is important not only for preventing hydrolysis but also because it enhances the rate of color formation. Ammonia also appears to play a role in the reaction rate and, in addition, it reduces the interference from copper. Cobalt is the most serious interference because its reaction with dimethylglyoxime is relatively slow under the experimental conditions used and, if significant amounts are present, the absorbance at 465 nm slowly increases with time. Amounts of cobalt less than 200 µg do not interfere in the determination of as little as 20 µg of nickel if the absorbance is measured within 15 minutes. When larger amounts of cobalt are present, the absorbance is measured at 465 and 410 nm and an appropriate correction is applied to the nickel results. The interference from copper is corrected for in a similar fashion. By performing absorbance measurements at these two wavelengths and utilizing the fact that formation of the cobalt complex is slow, appropriate corrections can be determined for both copper and cobalt. Conversely, these corrections can and have been used to estimate any copper, cobalt, or copper plus cobalt that may be present.

Iron and manganese in amounts greater than 1000 and 250 µg, respectively, in a final volume of 50 mL interfere by formation of precipitates. The volume of test solution must be sufficiently small to keep manganese below this level. The interferences of large amounts of iron is eliminated by extraction with a 1:1 mixture of methyl isobutyl ketone and amyl acetate.

To date, the persulfate-dimethylglyoxime procedure has been used for the spectrophotometric determination of nickel in more than 50 SRM's. Concentrations measured have ranged from 0.005 to 80 percent. The scope of the method and the agreement of the spectrophotometric results with the classical gravimetric values are illustrated by the data given in Table 13.

Table 13. Comparative Determinations of Nickel in Various SRM's by Gravimetry and by the Persulfate-Dimethylglyoxime Method [28].

SRM	Material	Ni, % Gravimetric	Ni, % Spectrometric	Difference
5j	Cast iron[a]	0.018	0.016	-0.002
153a	Co8-Mo9-W2-Cr4-V2 steel[a]	0.168	0.160	-0.008
37e	Sheet brass	0.53	0.52	-0.01
82b	Nickel-chromium cast iron[a]	1.22	1.22	0.00
346	Valve steel (Cr22-Ni4-Mn9)[a]	3.94	3.94	0.00
167	Cobalt-base alloy Co43-Mo4-Nb3-W4	20.65	20.57	-0.08
1193	High temperature alloy W 545	23.38	28.33	-0.05
	Permalloy	79.62	79.75	+0.13

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[a] Iron preextracted.

To improve precision and accuracy, differential measurements are recommended when more than 200 µg of nickel are present. Successful analyses have been performed using as much as 800 µg of nickel in a final volume of 50 mL (A ≅ 4) for the reference solution.

Unfortunately, IDMS values are not available for nickel in any of the above SRM's and the accuracy of the spectrophotometric results cannot be established unequivocally. Although the agreement between the gravimetric and spectrometric data are excellent, the possibility exists, however remote, that both techniques have the same bias. One set of analyses in which this situation occurred involved the determination of nickel in the 500 and 50-ppm trace elements in Glass SRM's (SRM 610 and 612, respectively). For the 500-ppm glass, the following average values were obtained: spectrophotometry, 451 ppm; polarography, 450 ppm, and IDMS, 458.7 ppm. Similarly, for the 50-ppm glass, the average values were 37.6, 37.8, and 38.8 ppm, respectively. A comparative plot of the spectrophotometric and IDMS results is shown in Figure 21.

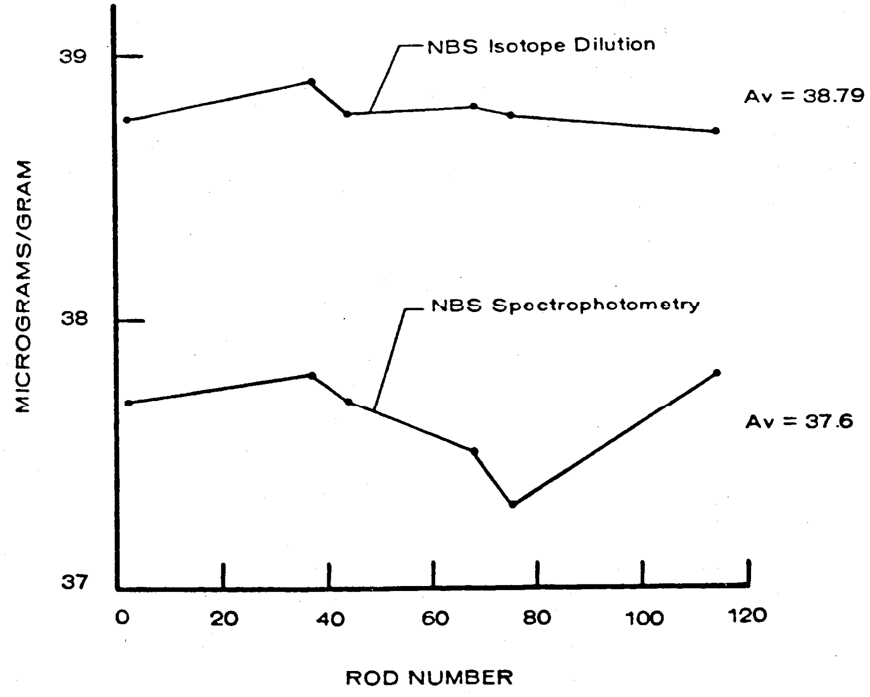

Figure 21. Comparison of spectrophotometric and IDMS results for nickel in NBS Trace-Elements in-Glass SRM 612.

Although the spectrophotometric and polarographic values were in agreement, both exhibited negative biases of almost two percent for the 500-ppm glass and 3 percent for the 50-ppm glass. In analyzing the possible sources of these biases, it is highly unlikely that the two techniques had inherent errors of these magnitudes in the measurement steps. Consequently, the low results can best be explained by the loss of nickel during sample dissolution. The base glass from which these SRM's were prepared contained approximately 12 percent CaO and included in the 61 doping elements were all of the rare earths. Both calcium and the rare earth elements form relatively insoluble fluoride salts. Since hydrofluoric acid was used for sample dissolution, it is highly possible that the losses resulted from some coprecipitation or adsorption of nickel on the insoluble fluoride residues.

7.3. Simultaneous Determination of Cobalt, Nickel, and Copper

One of the few spectrophotometric reagents that we have used successfully for simultaneous multielement analysis is 2,3-quinoxalinedithiol. In aqueous ethanol, cobalt (II), nickel (II), and copper (I) react instantaneously with this reagent at pH 2 to form intensely colored complexes with absorbance maxima at 510, 606 and 665, and 625 nm, respectively. At pH 6, the reaction of copper is virtually eliminated while the cobalt and nickel reactions exhibit only slight decreases in sensitivity. The absorbances of these complexes are additive for all combinations of these elements in which the individual concentrations are varied from 0.1 to 1 ppm. The wavelength differences in their absorbance maxima in conjunction with the sensitivity of the copper complex to pH and the additivity of the absorbances of the three complexes has been made the basis for the simultaneous method. The details of this method have been described previously [29]. In practice, two sample aliquots are required for analysis, one for color development at pH 2 and the other for color development at pH 6.

Iron, platinum, and palladium are serious interferences. For analysis of iron-base alloys, the iron is readily extracted with a 1:1 mixture of methyl isobutyl ketone and amyl acetate. Two extractions from 6 to 8 N hydrochloric acid are generally sufficient. The results obtainable for some typical SRM's is shown in Table 14.

Table 14. Simultaneous Determination of Cobalt, Nickel, and Copper in NBS SRM's.

SRM No.	Type	Certified Value, weight %	Found, weight %
462	Low alloy steel	Co 0.11	0.106
		Ni 0.70	0.687
		Cu 0.20	0.207
466	Ingot iron	Co 0.046	0.046
		Ni 0.051	0.051
		Cu 0.033	0.033
55e	Open-hearth iron	Co 0.007	0.0065
		Ni 0.038	0.037
		Cu 0.065	0.062
87a	Aluminum-silicon alloy	Co ...	<0.01
		Ni 0.57	0.568
		Cu 0.30	0.301

7.4. Copper

The simplest spectrophotometric analysis that we perform is the determination of copper as the dibenzyldithiocarbamate complex. The yellow-brown copper complex (λ_{max} = 435 nm) is formed and simultaneously extracted by equilibrating the sample solution for 1 to 2 minutes with a 0.01 percent solution of zinc dibenzyldithiocarbamate in either chloroform or carbon tetrachloride. The use of the colorless zinc chelate as the chromogenic reagent increases the selectivity of the method many common elements such as iron, chromium, or manganese form less stable complexes than zinc and do not undergo the exchange reaction. Selectivity is enhanced further by performing the extractions from 1 to 10 N hydrochloric or sulfuric acid. Under these conditions, bismuth and palladium are the only elements that might interfere by formation of similarly colored complexes. The method has been used most recently for the determination of copper in NBS botanical SRM's. A comparison of the spectrophotometric values and the isotope dilution-spark source mass spectrometric results is shown in

Table 15. The differences in the results obtained for copper by these two techniques are believed not to be significant.

Table 15. Comparison of Spectrophotometric and IDMS Results for Copper in NBS Botanical SRM's.

SRM No.	Material	Copper, ppm Spectrophotometric	Copper, ppm IDMS
1570	Spinach	12.0 ± 0.2[a]	11.5 ± 0.6[a]
1573	Tomato leaves	10.8 ± 0.2	10.2 ± 0.6
1575	Pine needles	2.9 ± 0.2	2.9 ± 0.4

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[a] 95% confidence interval.

7.5. Other Elements

Antimony, arsenic, nickel, cobalt, and copper are not the only elements that can be conveniently and accurately determined in a wide variety of matrices spectrophotometrically. The examples cited were selected because of the frequency of requests that we have for their determination and because of the long-term experience that we have had in applying these methods to the analysis of NBS SRM's. As the SRM program continues to expand from the traditional metal standards into such newer areas as clinical, environmental, and biological standards, the analytical needs also continue to change. If this paper were being written three years hence, the elements cited might more appropriately include selenium, lead, beryllium, molybdenum, iron, chromium, boron, titanium, zirconium, and zinc.

8. Future Developments in New Spectrophotometric Methodologies

Since Tschugaeff first described the synthesis and use of dimethylglyoxime for the detection of nickel in 1905, analysts have optimistically sought new and more sensitive and selective organic reagents for use in inorganic analysis. While many useful reagents and methods have evolved there have been no dramatic breakthroughs. Recently, perhaps the most outstanding approach of note for improving concomitantly sensitivity and selectivity has been the development of methods based on the formation of ternary complexes. Although the molar absorptivities of complexes used in inorganic analysis are typically in the range of 10,000 to 30,000 litre $mol^{-1}cm^{-1}$, Braude [30] has calculated, using molecular dimensions and absorption probability, that the theoretical maximum value of molar absorptivity is about 100,000 litre $mol^{-1}cm^{-1}$. In methods utilizing ternary complex formation, the element of interest reacts not with one ligand, but rather with two, that is, the brilliant green-hexachloroantimonate complex discussed previously. In this way it is possible for a larger and more strongly absorbing organic envelope to be placed around the element than is normally possible and consequently the sensitivity of the resulting ternary system is frequently superior. What is even more important is that the selectivity of ternary complex formation is likely to be much superior to its binary counterpart.

West [31,32] and coworkers have developed a number of useful methods based upon formation of highly colored and selective ternary complexes. The same workers have also advocated overcoming the sensitivity barrier through use of amplification procedures based upon allowing the element determined to react in a catalytic fashion or by exploiting a favorable stoichiometric relation between the element measured and the element sought. Because of their favorable characteristics and the fact that they lend themselves well to systematic study and application, the areas involving ternary complex formation and amplification techniques appear to merit considerable future attention.

Probably the most exciting and challenging development at the moment in molecular absorption spectrometry is the possibility of using high performance liquid chromatography (HPLC) to separate mixtures of metal chelates prior to their subsequent quantitation through dynamic coupling of the chromatograph to a variable wavelength, ultraviolet-visible spectrophotometer. While HPLC has experienced a tremendous growth in the last seven or eight years, almost all of the work has been in organic rather than inorganic analysis. The worldwide use of this technique for separating complex mixtures of polynuclear aromatic hydrocarbons, drugs, pesticide residues, and the like is now commonplace. Only a few papers have appeared dealing with the HPLC separation of metal chelates and none have been used for practical analyses. Included among these is the paper by Huber et al., [33], the several publications by Ballschmiter et al., [34-36], the paper by Uden and Bigley [37], and, most recently, a paper by Schwedt [38].

Our current research interests are centered on the use of nonselective chelating agents for group solvent extractions of the transition and heavy metals and their subsequent HPLC separation and quantitation by spectrophotometric detection. Reagents under study include α-pyridylazonaphthol, diethyldithiocarbamate, and dibenzyldithiocarbamate. The separation of mixtures of metal complexes with each of these reagents is being examined on several types of column packings and for various compositions of the mobile phase. In general, separations on silica have been superior to those obtained on bonded-phase $-NH_2$, $-CN$, and $-C_{18}$ packings. A typical chromatogram of a mixture of copper, cobalt, and nickel dibenzyldithiocarbamates is shown in Figure 22. Although data are shown only for three elements, it is apparent that half a dozen or more elements can be separated with baseline resolution and determined in a 30-minute run provided their complexes have appropriate retention times. Presently, the major problem is not obtaining the desired separations but rather preventing on-column degradation of the metal complexes. Silica exhibits quite anomalous behavior in this respect. For example, the degradation of nickel diethyldithiocarbamate on 5 to 10 μm irregular silica is quite extensive, whereas, on spherical silica of the same particle size, degradation is drastically reduced [39]. Obviously much work remains to be done to understand and control the various interactions occurring on the column. Provided the problem of degradation can be surmounted, it is refreshing to believe that high performance liquid chromatography in conjunction with spectrophotometric detection can become a very practical and inexpensive multielement technique. The traditional requirement that an element can be determined accurately in a complex matrix by absorption spectrophotometry only when it has been separated from all interfering elements is still present in the proposed technique. The difference is that the HPLC operation provides quick and efficient separations instrumentally. Our optimism about the future of the technique is augmented further by its potential use in metallo-organic speciation studies and by the fact that the principles of isotope dilution can conceivably be applied for the determination of those elements having sufficient differences in isotopic mass and abundance.

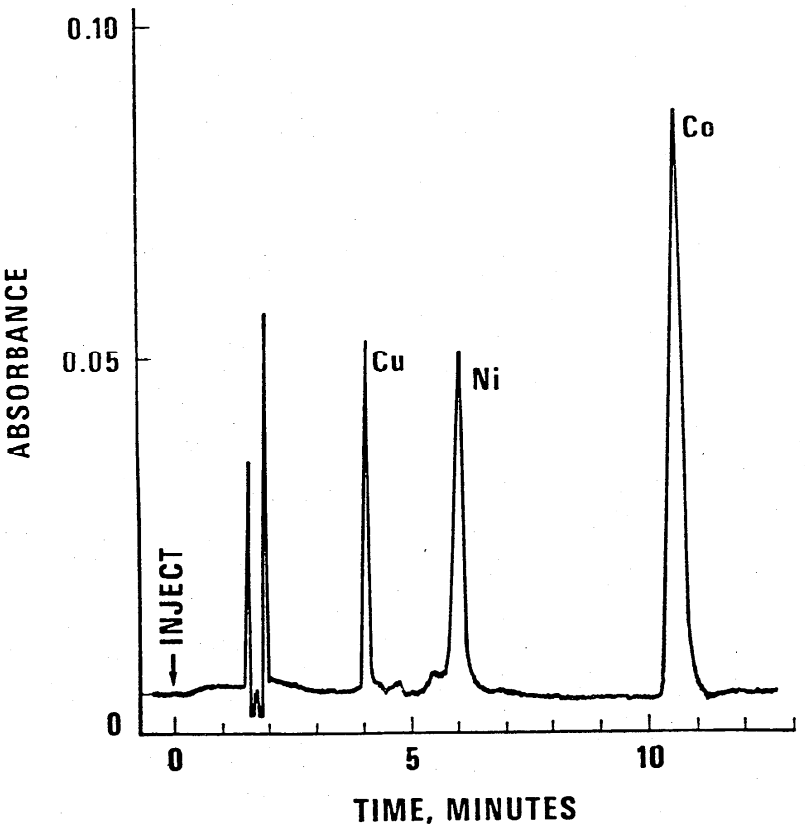

Figure 22. High-performance liquid chromatographic separation and spectrophotometric detection of a mixture of copper, cobalt, and nickel dibenzyldithiocarbamate chelates.

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APPENDICES

These appendices contain reproductions of several publications which should provide further and more detailed information on the subjects discussed in this work. Copies of the certificates for the SRM's described in this publication are also included in these appendices.

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Comments on Spectrometry Nomenclature

Sir: As an optical physicist who collaborates with analytical chemists in the development and use of absorption and luminescence spectrometers, I have become increasingly aware of the fact that our vocabularies are not always the same. Although some of these differences have been pointed out before (*1*) it seems worthwhile to familiarize the readers of this journal with the currently prevailing terminology of physical optics. Perhaps the following comments may be helpful to editors, authors, and nomenclature committees in establishing a common and consistent terminology that can be used in all branches of spectrometry and would save readers a good deal of puzzlement and reading between lines. The main purpose of these comments is to emphasize the need for interdisciplinary efforts in defining spectrometry nomenclature without perpetuating the inconsistencies that exist at present. They do not represent an official position of the National Bureau of Standards.

The basic concept of modern optical terminology (*2, 3*) is to combine nouns and adjectives in order to describe quantities and properties as explicitly as necessary, rather than using glossaries of short names for them. This "Chinese Restaurant" method of nomenclature (*4*) offers flexibility in introducing new terms, and has recently been extended to include the photon quantities used in photochemistry and similar disciplines dealing with the interaction of light and matter (*5*).

The basic list of nouns describing the transport of energy according to the laws of geometrical optics is:

Energy, Q.
Energy density, $u = \mathrm{d}Q/\mathrm{d}V$. Energy per unit volume.
Flux, $\Phi = \mathrm{d}Q/\mathrm{d}t$. Time rate of energy flow.
Flux density, $W = \mathrm{d}\Phi/\mathrm{d}A$. Flux per unit area.
Intensity, $I = \mathrm{d}\Phi/\mathrm{d}\Omega$. Flux per unit solid angle.
Sterance, $L = \mathrm{d}^2\Phi/\mathrm{d}(A\cos\theta)\mathrm{d}\Omega$. Flux per unit projected area and unit solid angle.

(A number of alternative and additional terms have been proposed. For example: *pointance* to replace the continually misused term *intensity; incidance* and *exitance*, or *incident areance* and (*transmitted, emitted*, etc.) *areance* for the flux densities arriving at and leaving a surface; *sterisent* for the sterance generated per unit path length by emission or scattering into the direction of propagation; and *fluence* for the surface energy density incident upon a volume element irradiated from within a large solid angle of rays (F. E. Nicodemus, private communication). As these have not yet been accepted generally, they were not included here. However, the new term *sterance* was included as it avoids the misnomer *photon radiance* that has appeared in papers on luminescence spectrometry.)

These nouns are modified by the adjective *radiant*, and the above symbols are written with a subscript 'e' (for energy), when radiometric units are used. The modifiers *luminous* and *photon* are used in conjunction with subscripts 'v' (for visual) and 'p' (for photon) to indicate the use of photometric and photon units, respectively. For example:

Radiant energy density, u_e[J m^{-3}].
Luminous intensity, I_v[lm sr^{-1}].
Photon sterance, L_p[E m^{-2} sr^{-1} s^{-1}].

(The basic unit for photon energy used here is the *einstein* [E], defined as one mole of photons. It is not an SI unit, but is used so extensively in photochemistry and photobiology that its acceptance as a supplementary SI unit may be desirable.)

The additional modifier *spectral* and subscripts λ and σ are used to denote derivatives of radiometric and photon quantities with respect to wavelength and wavenumber, respectively. Thus:

Spectral radiant energy density, $u_{e,\lambda} = \mathrm{d}u_e/\mathrm{d}\lambda$ [J m^{-4}].
Radiant energy density per unit wavelength interval.
Spectral photon intensity, $I_{p,\sigma} = \mathrm{d}I_p/\mathrm{d}\sigma$ [E sr^{-1} s^{-1} m].
Photon intensity per unit wavenumber interval.

Any of these can be modified further; such as: *fluorescence photon flux, transmitted spectral radiant flux density*, or *absorbed luminous energy*. Usually, it is possible to drop most of the modifying adjectives as well as the subscripts e, v, or p, once the context has been clearly established or whenever a distinction is not necessary.

The same method of nomenclature also provides a simple and logical way of specifying the precise meaning of the quantities and material properties commonly measured in analytical spectrometry. Thus *radiant absorptance* α_e and *photon absorptance* α_p should be used for the ratios of the radiant or photon fluxes absorbed by a sample to those incident upon it, when measured with a large bandwidth so that these two ratios are not the same. On the other hand, *spectral absorptance* $\alpha(\lambda)$ is sufficiently accurate in the case of measurements made with a small bandwidth. Similarly, luminescence yields (the ratios of the radiant or photon fluxes emitted by a sample to those absorbed by it) should be designated as *radiant yield* η_e or *photon yield* η_p (not *energy yield* or *quantum yield*). In this case, *spectral radiant yield*, $\eta_{e,\sigma} = \mathrm{d}\eta_e/\mathrm{d}\sigma$, and *spectral photon yield*, $\eta_{p,\sigma} = \mathrm{d}\eta_p/\mathrm{d}\sigma$, are different quantities even in the limit of infinitely narrow bandwidths, and thus should be referred to by these names.

[The dependence on wavelength or wavenumber is indicated by a subscript ($\eta_{p,\sigma}$) when the spectral distribution is defined as a derivative, but in functional form [$\alpha(\lambda)$] in the case of spectral distributions that are not derivatives. This is an important distinction that must also be borne in mind in the presentation of spectra. The positions of the peaks and valleys in derivative spectra depend on the units used, so that these should not be published in the form of a single graph with dual scales (such as wavelength and wavenumber, or spectral radiant and photon yield).]

In addition to this general description of current optical terminology, the following comments are made in direct reference to the nomenclature list in the December 1975 issue of *Analytical Chemistry* (*6*).

1) This list cautions not to use *optical density* instead of *absorbance*, but defines the latter as "the logarithm to the base ten of the reciprocal of transmittance"—which is the definition of optical density. *Internal transmittance* should have been substituted for *transmittance* in this def-

inition of absorbance. The International Lighting Vocabulary suggests *transmission density* and *internal transmission density* instead of optical density and absorbance. This is more determinative, and also fits into a general scheme where *reflection density* is defined as the negative logarithm to the base ten of reflectance. On the other hand, transmission and reflection density are operational quantities that merely express measured data on a logarithmic scale, whereas absorbance is directly related to molecular constants through the Lambert–Beer and Strickler–Berg equations. Thus, in my opinion, it should be retained as a separate term. I believe that all confusion would be avoided by defining:

Transmittance, τ. Ratio of the flux transmitted by a sample to the flux incident upon it.

Internal transmittance, τ_i. Transmittance exclusive of losses at boundary surfaces and effects of interreflections between them.

Transmittance density, $D = -\log_{10}\tau$. Negative logarithm to base ten of transmittance.

Absorbance (*Internal transmission density*), $A = -\log_{10}\tau_i$. Negative logarithm to base ten of internal transmittance.

2) The definitions of *absorptivity* in chemistry and physics are not the same. In chemistry, it means absor*b*ance per unit path length and unit concentration (A/bc), whereas the International Lighting Vocabulary (*2*) defines it as internal absor*pt*ance per unit path length ($d\alpha_i/db$). Similarly, it specifies *transmissivity* as internal transmittance per unit path length and *reflectivity* as the reflectance of a thick layer (so that a further increase in thickness will no longer change its value). Generally, terms ending in *-ance* represent sample properties, whereas terms ending in *-ivity* denote material properties that are independent of sample geometry. The quantity A/bc does not fall in this latter category as it is also independent of sample concentration. The International Union for Pure and Applied Chemistry, apparently aware of this discrepancy, has suggested calling it *absorption coefficient*. However, as this name has been given different and mutually inconsistent meanings in the past, a less ambiguous word—perhaps *specific absorbance*—would have been better. Therefore it is proposed to define:

Absorptivity, $a = d\alpha_i/db$ [m^{-1}]. Internal absorptance per unit path length.

Specific absorbance, $\epsilon = A/bc$ [$\mathrm{kg}^{-1}\ \mathrm{m}^2$]. Absorbance per unit path length and unit concentration.

Specific molar absorbance, $\epsilon_m = A/bc_m$ [$\mathrm{mol}^{-1}\ \mathrm{m}^2$]. Absorbance per unit path length and unit molar concentration.

3) The December 1975 nomenclature list also implies that the Beer–Lambert laws are the same. The correct definitions are:

Beer's law: Absorbance is proportional to concentration.

Lambert's law: Absorbance is proportional to path length. Also called *Bouguer's law*.

4) The definition of the *ångström* unit in terms of the red line of cadmium has been abrogated several years ago. It is now defined as 10^{-10} m, exactly. However, the ångström is not an SI unit, and has been sanctioned by the International Committee on Weights and Measures only as a supplementary unit that will eventually be abandoned (*7*). Therefore, authors should be encouraged to use *micrometers* or *nanometers*.

5) The December 1975 list defines *spectrometry* as the "measurement of spectra", but restricts the meaning of *spectrometer* to "instrument with an entrance slit, a dispersing device, and with one or more exit slits . . .". As this excludes non-dispersive and slit-less instruments, such as Fourier and Girard spectrometers, it would be more consistent to call any instrument used for spectrometry a *spectrometer*. Therefore:

Spectrometer: Instrument for the measurement of spectra.

The general term for instruments that measure spectral distributions of radiometric quantities is *spectroradiometer*. However, as this implies measurements in absolute units, *spectrometer* is a better term for the simpler, usually ratio-forming instruments used in analytical spectrometry. Thus, a *spectrophotometer* could also be called *absorption spectrometer*, and *fluoroescence spectrometer* would end the controversy of *spectrofluorimeter* vs. *spectrofluorometer*.

(Strictly speaking, the word *spectrophotometer* is a misnomer. A *photometer* is an instrument that measures luminous flux in lumens. Since the adjective *luminous* implies the integral effect of visual radiation as perceived by the human eye, the spectral analysis of luminous flux has no physical meaning. However, in view of the firmly established meaning of *spectrophotometer*, it is not suggested to change it, although a scanning of the recent literature shows an increasing usage of *absorption spectrometer*.)

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An Accurate Spectrophotometer for Measuring the Transmittance of Solid and Liquid Materials

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The optical transmittance of solids and liquids as well as the molar absorptivity of various chemical species are parameters of fundamental significance in characterizing these materials. Meaningful transmittance data can be obtained only when the measurements are performed with well-known accuracy and precision. To perform such measurements, a high accuracy spectrophotometer was designed and assembled at NBS, Analytical Chemistry Division, and will be described in this paper. This single-beam instrument is composed of a constant radiation source, a monochromator, a sample carriage, an integrating sphere-photomultiplier assembly followed by appropriate electronics, and a read out system consisting of a digital voltmeter and a computer data acquisition and handling provision. The accuracy of transmittance measurements is determined by the light-addition principle used in conjunction with a two-aperture arrangement. The spectrophotometer can be used in manual or automatic modes of operation. A detailed discussion of the data obtained with this instrument, used in both modes, will be presented together with its application to the certification of solid and liquid Standard Reference Materials for checking the photometric scales of conventional spectrophotometers.

Key words: Absorbance; automation of accurate spectrophotometer; instrumentation, spectrophotometric; spectrophotometry, high accuracy; standard reference material in spectrophotometry; transmittance.

1. Introduction

Optical transmittance is due to an intrinsic property of matter and characterizes a particular transparent material. Since this parameter is not known a priori, it must be determined by experimental procedures.

True transmittance values can be obtained only by using accurate measuring techniques and by taking into consideration all factors which can affect and distort the data.[1]

Transmittance is the ratio of two radiation flux intensities. It is therefore necessary that the photometric scale of the spectrophotometer used to perform the measurements be accurate. The transmittance of a particular material is also a function of wavelength: hence the wavelength scale of the monochromator should also be accurate, and appropriate spectral bandpasses should be used. The measurements should be made using collimated radiations. Such radiations define unambiguously the actual path length through the transmitting medium, the reflection losses, and eliminate the effects of polarized radiations that are produced at the surface of the sample. Other important factors which must be considered are: homogeneity and stability of the sample, radiation scatter inside the sample, interference phenomena, stray radiation, polarization, fluorescence, temperature, particulate matter, and surface conditions. Since transmittance measurements depend on a diversity of factors, meaningful values can be obtained only by defining the experimental conditions for obtaining transmittance data [1, 2].[2] Spectrophotometers are used to perform two types of measurements:

(1) Quantitative determination of chemical species using the relation between optical transmission of the material, and the concentration as a measuring parameter. Under these circumstances, the photometric scale

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[1] The optical transmittance of a solid material includes the reflection losses which occur at the air-solid interface.

The internal transmittance is defined as the transmittance of the material corrected for reflection losses (2). This internal transmittance can be calculated in principle from the transmittance by using the well known Fresnel equations (1, pp. 98 to 100).

For collimated radiation the reflectance R, for a material with an index of refraction, n, and an absorptivity, a, at wavelength, λ, is given through:

$$R_\lambda = \frac{(n_\lambda - 1)^2 + n_\lambda^2 a_\lambda^2}{(n_\lambda + 1)^2 + n_\lambda^2 a_\lambda^2}$$

For a nonabsorbing material and collimated radiation:

$$R_\lambda = \frac{(n_\lambda - 1)^2}{(n_\lambda + 1)^2}$$

For glass, n is approximately 1.5 in the visible region of the spectrum, and R will be about 4 percent at every air-glass interface.

When noncollimated radiation is used:

$$R_\lambda^\perp = \frac{\sin^2 (\alpha - \beta)_\lambda}{\sin^2 (\alpha + \beta)_\lambda}$$

for perpendicular polarized radiation, and

$$R_\lambda^\parallel = \frac{\tan^2 (\alpha - \beta)_\lambda}{\tan^2 (\alpha + \beta)_\lambda}$$

for parallel polarized radiation, where α and β are the angles of incidence and refraction, respectively.

In collimated radiation and in air, $\alpha = \beta = 0$ and $R^\perp = R^\parallel = R$.

[2] Figures in brackets indicate the literature references at the end of this paper.

of the spectrophotometer is calibrated in meaningful units, using a series of reference solutions having known concentrations of the species to be determined, rather than values of optical transmittance.

The accuracy of the measurements is related solely to the accuracy with which the concentration of the reference solutions is known and to the precision (stability, sensitivity, reproducibility) of the spectrophotometric method and instrument used. The accuracy of the photometric scale per se, is not a critical factor in such measurements.

The precision, stability, and reproducibility of the instrument can be checked before each series of measurements by careful use of solid or liquid reference filters having well established transmittance values.

(2) Determination of the optical transmission characteristics of solid or liquid materials, and the determination of molar absorptivities of chemical compounds. In both cases the accuracy of the photometric scale of the measuring instrument, among other things, is essential to provide true values. Ways to establish and check this important parameter are critically needed.

Since conventional spectrophotometers do not provide means to check photometric accuracy or to evaluate the possible sources of systematic errors, it was decided in 1969 to design and construct a research spectrophotometer on which transmittance measurements could be performed with well defined accuracy. Such an instrument would be used to determine optical transmittance of selected solids and liquids at various wavelengths. These materials can be used as standard reference materials (SRM's) to check the accuracy of the photometric scale of conventional spectrophotometers. The same certified SRM's could likewise be used to monitor the precision, stability, and reproducibility of those instruments [3, 4].

After a comprehensive examination of the literature in this field [5 to 34] arranged in chronological order, an instrument was developed which is similar in principle to the instrument at the National Physical Laboratory (NPL), Teddington, England, where a long tradition of high accuracy spectrophotometry exists. The instrument described in this work performs measurements of radiant energy in the visible and ultraviolet region of the spectrum, with well established and high photometric accuracy. Transmittance measurements on solids and liquids can be made with this instrument using collimated as well as noncollimated beam geometry. The wavelength accuracy and spectral bandpass achievable are adequate to avoid degradation of photometric accuracy, and the other interferences mentioned have been given careful consideration, and, in most cases, have been assessed quantitatively.

The transmittance measurements on the optically neutral glass filters discussed in this work have been made with a noncollimated beam geometry corresponding to an aperture of about f:10. The image of the exit slit of the monochromator (8 mm x 0.5 mm) was produced at the center of the entrance face of the filter. All measurements have been made against air for the nonattenuated radiation flux, and no correction for reflection losses was made. Transmittance measurements made with noncollimated radiation by projecting the image of the exit slit of the monochromator on the entrance face of the sample using an opening of f:10 (total angle of about 7° or 8°), may differ by several parts in 10^4 of the value when compared with similar measurements made with collimated radiations, as indicated in this Journal by K. Mielenz.

Noncollimated beam geometry was applied in this work to approach the measuring conditions used in most of the conventional spectrophotometers which are available today. A brief description of this instrument was given earlier in reference [3].

II. Description of the Instrument[3]

The high accuracy spectrophotometer, completed and tested in 1970, is a single beam instrument which contains the following components: (a) a constant radiation source, (b) a monochromator, (c) a sample holder, (d) a system to check the accuracy of the photometric measurements, (e) an integrating sphere attached to a photomultiplier-digital voltmeter unit, and (f) the data presentation system. Figure 1 illustrates schematically the arrangement of these various components. A circular neutral wedge is placed after the light source to select various levels of radiation intensities required for measurements. A description of the components is presented in the following sections.

a. **The Radiation Source.** Since the instrument is a single-beam type, it is essential that the radiation source be constant and homogeneous. Additional desirable conditions are: capability of monitoring the current supplied to the source and radiation similar to that from a Planckian radiator. The source is similar in design to that developed and used at NBS by H. J. Kostkowski and R. D. Lee of the Institute for Basic Standards. This source was duplicated in our instrument with the kind assistance of its developers.

The source is used in the spectral range 360 nm to 800 nm and consists of a tungsten incandescent filament lamp with a tungsten ribbon 8 mm long by 2 mm wide. The connections to the lamp terminals are soldered to minimize contact problems (see fig. 6). The direct current required to operate this lamp at approximately 3000 K is 18 A across a 6 V drop; our source is operated at 5 V and 15 A. The d.c. power supply is capable of delivering 15 V and 50 A, and can be operated in constant current or constant voltage modes. To achieve the constant current mode an external sensing resistor of 0.1Ω and 50 A and a current control circuit are placed in series with the power supply. A feedback voltage across this resistor is connected to the sensing system. The character

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[3] The commercial instruments and parts used in the construction of the spectrophotometer are identified in the addendum.

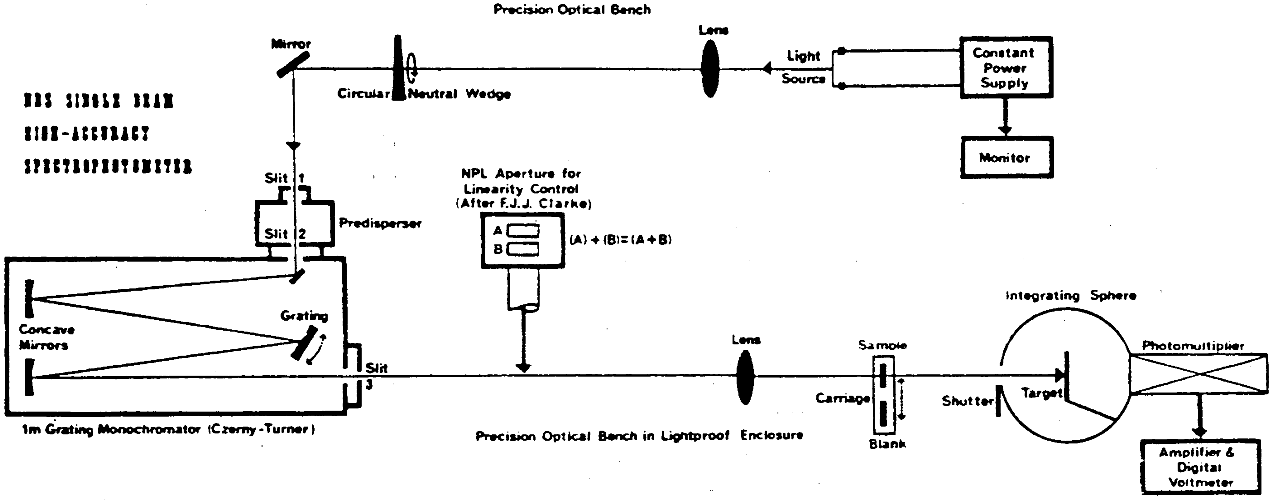


FIGURE 1. Principle of the single beam high accuracy spectrophotometer. The aperture unit is placed on the optical bench only when linearity measurements are performed.

istic of this function is the ability to automatically change its output voltage to maintain a constant current to the load resistor, which, in our case, is the lamp source. The nominal current regulation obtained is better than 0.01 percent, and the stability over an 8 hour period, at constant load temperature, is better than 0.02 percent. The stability of the current delivered to the lamp is monitored with a high accuracy potentiometer used in conjunction with a null meter. This meter is sensitive to variations in the current supplied to the lamp from 1 part in 1000 to 1 part in 1,000,000 per division (fig. 1 and fig. 14). The potentiometer is connected to the current source across a resistor (0.01Ω and 100 A) placed in series with the lamp.

The demagnified (2 to 1) image of the ribbon filament is projected on the entrance slit of the predisperser by a fused quartz (nonfluorescent SiO_2) lens whose focal distance is 254 mm and diameter is 44 mm. This and the other lenses used in the optical system, were calculated by K. Mielenz of the Institute for Basic Standards at NBS. The lenses are mounted in carriers which permit orientation in any position. A circular neutral wedge is placed between the light source and the predisperser. This wedge, evaporated inconel on a fused quartz disc (150 mm diam), is linear in density and provides a light attenuation of 100 to 1. The wedge is motor driven (1 rev. per s) to select proper radiation intensity levels as required by the measurements (figs. 2, 3, and 4). The radiation source used for measurements in the ultraviolet region to 275 nm is a single coil tungsten-bromine incandescent lamp (fig. 5) supplied by an adequate power source; below 275 nm, a deuterium discharge lamp is contemplated.

b. **The Monochromator.** The monochromator is a 1-m Czerny-Turner type grating instrument with a dispersion of 0.8 nm/mm. The flat grating has 1200 grooves per mm covering a surface of 100 x 100 mm. The monochromator is provided with a predispersing attachment to reduce the stray light (fig. 3). This pre-

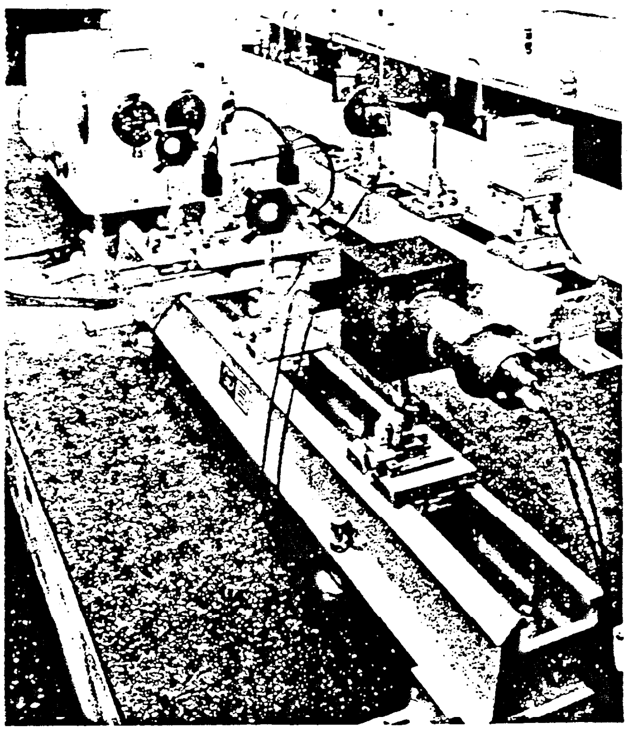


FIGURE 2. General views of the spectrophotometer. Rear: optical bench carrying the tungsten-halogen radiation source used for checking the alignment of optical components, followed by a quartz lens, the circular quartz neutral wedge, and a flat mirror. Left: the 1-m Czerny-Turner grating monochromator (the predisperser is not illustrated here). Front: optical bench carrying a quartz lens, the single-sample and blank carriage, a second quartz lens, and the integrating sphere with the photomultiplier housing.

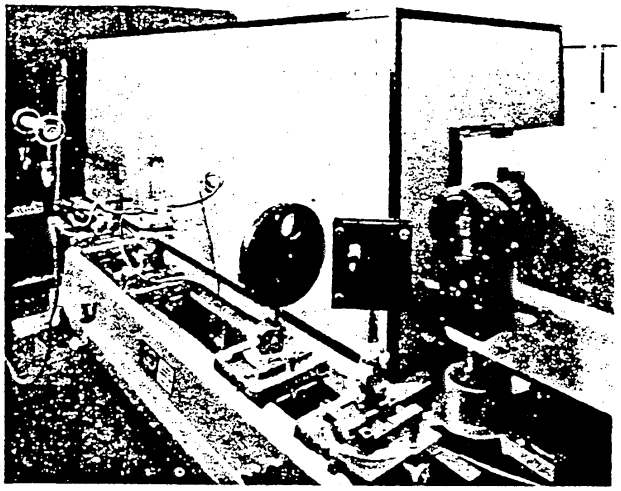


FIGURE 3. Close view of the tungsten ribbon filament lamp on its adjustable holder, followed by the quartz lens—circular neutral wedge assembly, and by the flat mirror in its adjustable holder. The 30° quartz prism Littrow-type predisperser is located at the entrance slit of the 1-m grating monochromator. Extreme left: neon gas laser used to check the optical alignment, and mercury discharge lamp for wavelength calibration. When in use, the tungsten ribbon lamp is surrounded by an enclosure with black walls (50 cm x 50 cm x 70 cm high). Rear: enclosure containing the optical units illustrated in figure 2.

disperser is a small quartz prism monochromator connected to the scanning system of the 1-m instrument. A wavelength counter permits readings to 0.1 nm and the scanning speed can be varied from 0.05 nm to 200 nm/min by a 12 speed synchronous electric motor.

The optical components are placed on precision lathe bed type optical benches which are 160 and 120 cm long, and are equipped with appropriate carriers provided with *x-y-z* adjustments.

c. **Sample Carrying Systems.** The spectrophotometer is provided with two sample carrying systems. One system measures one sample and its blank, while the other system permits sequential measurements for seven samples and eight reference reading positions against air, and can be operated manually or automatically through a computer interfaced with the instrument.

The single sample carrying unit consists of a platform provided with two vertical holders which can accept ¾-in (14 mm) rods and a variety of sample supports (fig. 2). These holders can be moved laterally through a rack and pinion arrangement. The platform is mounted on 4 ball bushings which ride on two horizontal rods and can be moved pneumatically across the optical axis. The pneumatic operation was recommended by G. E. Moore and J. T. Sterling of the Institute for Materials Research at NBS and by L. Owen, a guest worker at NBS. The travel distance is 8 in (20 cm) and the linear movement is smooth; the position of the platform and the sample in and out of the optical beam, can be reproduced within 0.025 mm. This unit is illustrated in figure 2 and is located between the two quartz lenses. The sample holder is designed to accept conventional solid or liquid filter holders which fit most spectrophotometers. These holders are provided with a thermostating jacket, and can be rotated in the horizontal plane through a 10 cm diameter rotating table.

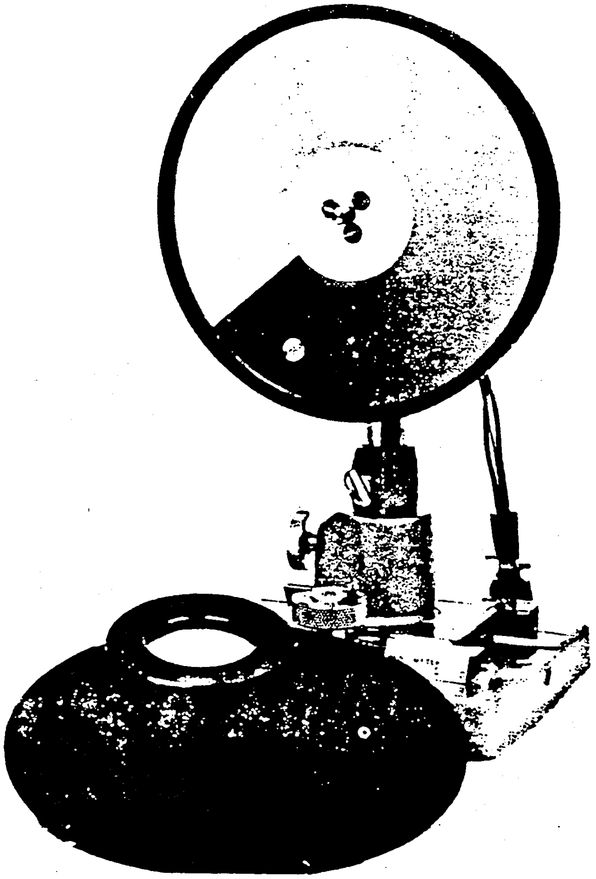


FIGURE 4. Close view of the circular neutral wedge. The front plate which carries the fused silica lens was removed to show the fused silica disc with the evaporated metal layer.

A filter holder which permits the rotation and scanning of the sample in the *x–y* direction is also available (fig. 7). It is provided with micrometer screws having a total linear motion of 25 mm with 0.01 mm per division. The seven-sample carrying unit is illustrated in figures 8 and 9 and consists of a semicircular aluminum-alloy plate placed horizontally on an appropriate carrier on the optical bench along the optical axis. This plate, which is 32 cm in diameter and 2.5 cm thick, can be rotated clockwise through a pneumatically operated precision ratchet system in increments of 12°. The stepwise rotation utilizes a solenoid valve which is operated electrically by a switch located outside the enclosure. This switch can be operated manually or automatically by computer (fig. 14).

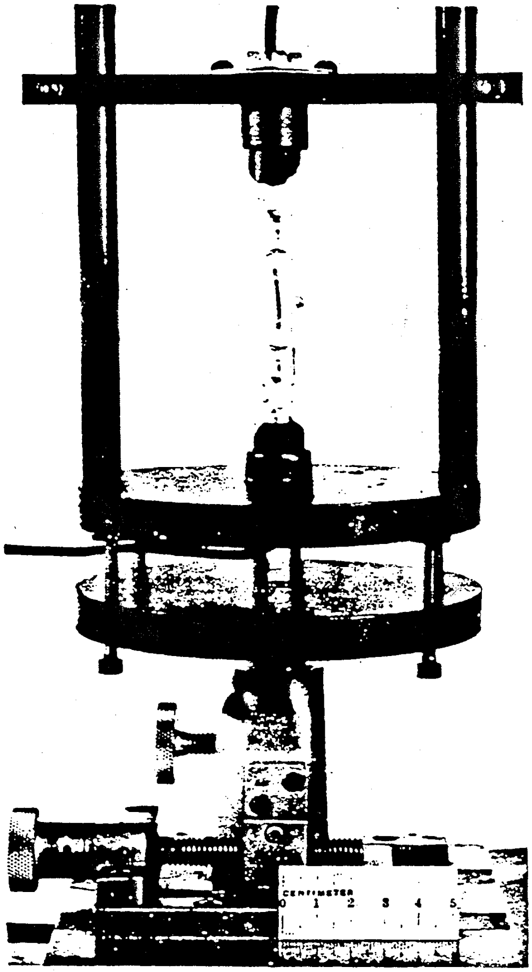


FIGURE 5. Single coil tungsten halogen lamp in the adjustable holder.

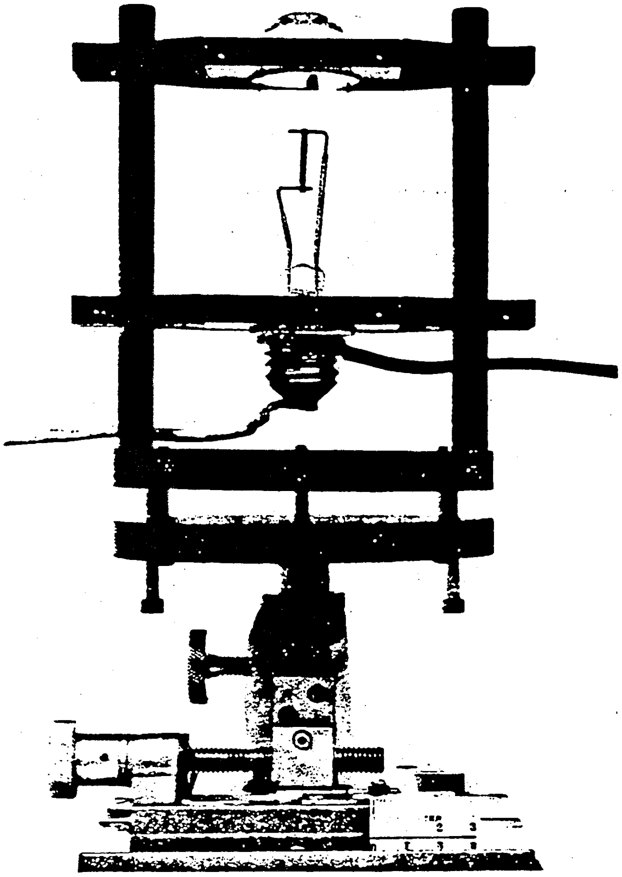


FIGURE 6. The tungsten ribbon filament lamp in the newly designed adjustable holder. The platform which carries the lamp is similar to that described in figure 5 and can be oriented in the horizontal plane through the six screws spaced around the edges of the platform at 60° intervals. Three screws push the platform while the other three pull.

The current-supplying wires are soldered directly to the lamp terminals to eliminate contact problems.

The semicircular plate carries seven sample holders similar to those used for the single sample system described earlier. The holders are placed at 24° intervals and are separated by blank spacings. About 1 atm of air pressure is used to operate the plate and the rotation is set at 2 s per 12° step when the automatic computer operating mode is used.

d. System to Check the Accuracy of the Photometric Reading. Since the high accuracy spectrophotometer is single beam, accurate photometric data are obtained when there is a linear relation between the measured radiation flux and the corresponding response of the photodetector.

Linearity of photodetectors can be measured by several means: the inverse square law [7, 15]; the use of optical elements having a known transmittance which can be determined by other means [17] and the light addition principle of Elster and Geitel using a plurality of light sources [5, 6, 8, 9, 10, 13, 18, 19, 20, 28, 31, 33, 34] or multiple apertures [11, 12, 14, 16, 21, 23, 25, 26, 27, 30]. A novel approach to the problem of accurate photometric measurements was described by O. C. Jones and F. J. J. Clarke [24, 29] and by F. Desvignes and J. Ohnet [32]. A critical discussion of some aspects of accurate spectrophotometry will be found in an NBS manuscript by Gibson and associates [22]. The radiation addition principle, using two apertures with one source of radiation, was chosen for our work. The aperture method for checking the linearity of photometric data was in use at the National Physical Laboratory from about 1930 onwards, and one form of it was described by Preston and Cuckow [11] in conjunction with a single beam spectrophotometer, using a five aperture screen. One year later, Buchmüller and König [12] described and used a two aperture unit. At NBS, Barbrow [14] used a 10 aperture arrangement, while Harding [16] and Cordle and Habell [25] at NPL described a two aperture system. Multiapertures were used by Hoppmann [21], Bischoff [23], Sanders [26] and Nonaka and Kashima [27]. Finally, Clarke [30]

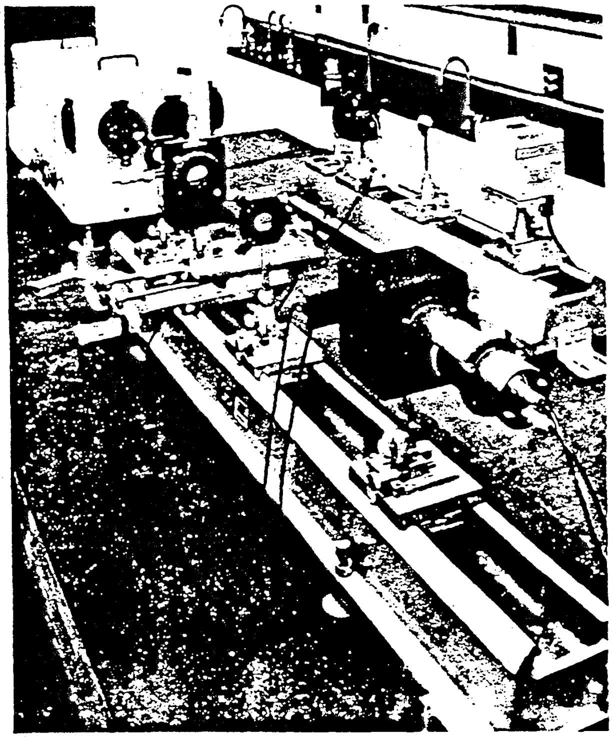


FIGURE 7. Same as figure 2 except for the sample holder which in this case is capable of rotating the sample 360° and to displace it in the x-y direction through the micrometer screws.

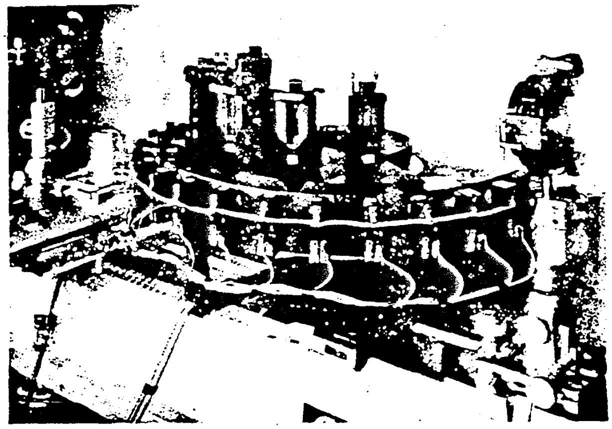


FIGURE 8. Circular platform carrying seven filter holders. The 15 position switches (7 sample positions and 8 blank positions) are visible along with the two quartz lenses. The exit slit of the monochromator is at left.

discussed in detail the use of a two aperture system to check the accuracy of photometric data obtained on the spectrophotometer at NPL. It is this two aperture system which is used at NBS.

The two aperture unit consists of a metal plate (130 mm by 100 mm) containing two rectangular windows, A and B, (20 mm by 8 mm) located one above the other (figs. 10, 11, 12). Each aperture can be closed by a light-tight shutter which is operated pneumatically by remote control (fig. 14). The aperture plate is placed in the optical path after the exit slit of the monochromator and *within* the optical solid angle of the instrument. The image of the apertures is then projected on the target of the integrating sphere. A fused quartz lens with a focal distance of 190 mm and a diameter of 60 mm is used for this purpose. The arrangement is illustrated in figure 10. No optical element

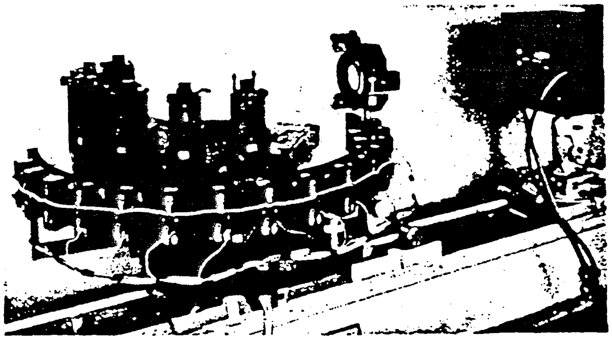


FIGURE 9. Same as figure 8. The pneumatic cylinder which rotates the circular platform through a ratchet mechanism is visible at the rear of the platform. The integrating sphere with its pneumatic shutter is seen at right.

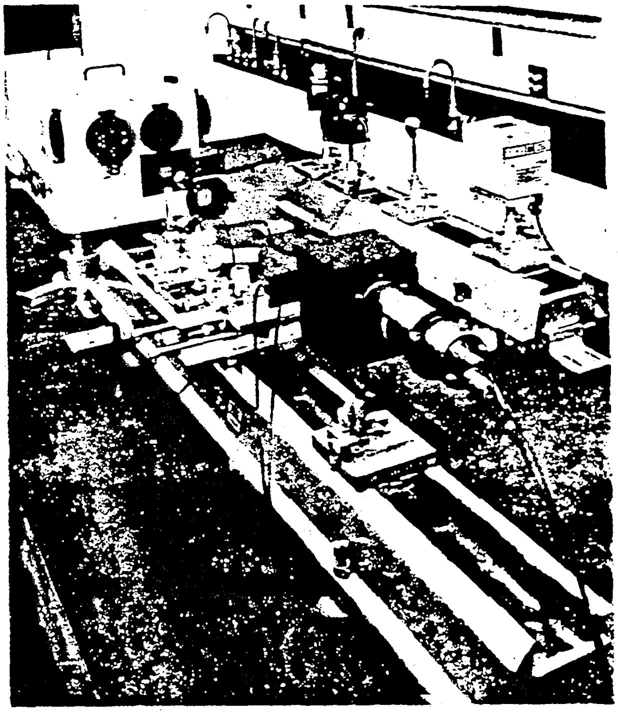


FIGURE 10. Same as figure 2. In this case the dual-aperture unit for linearity control is located on the optical bench after the exit slit of the monochromator.

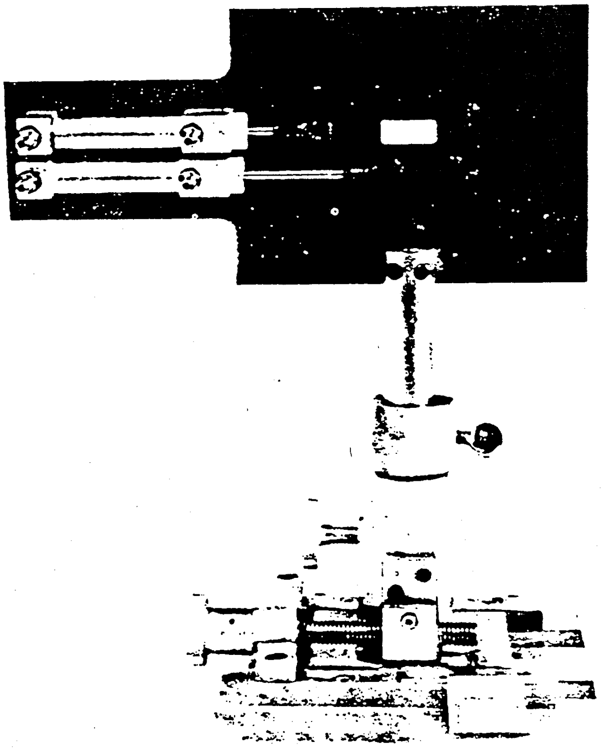


FIGURE 11. Detail of the dual-aperture unit showing its construction and the pneumatic system which operates the two shutters. One aperture is open, the other is closed.

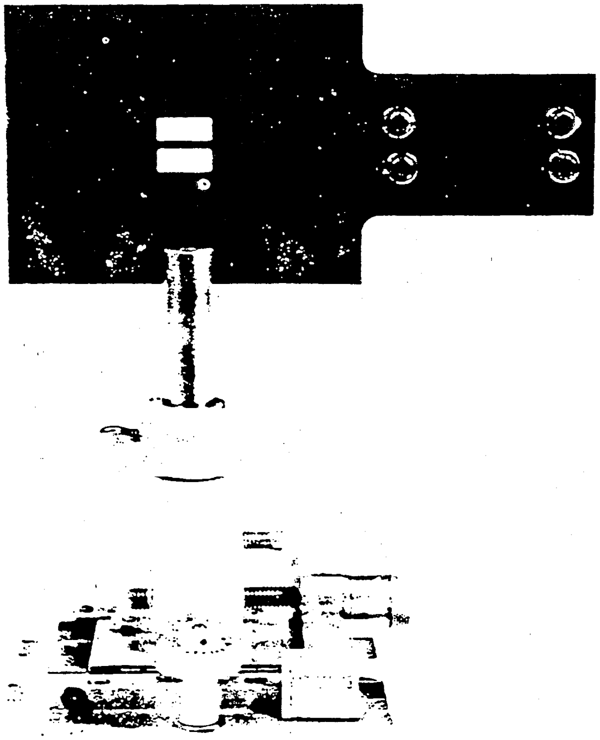


FIGURE 12. Front view of the dual-aperture unit, with both apertures open.

should be placed between the aperture plate and the monochromator. The linearity check consists of measuring the photocurrent produced when aperture A is open then closed, and then aperture B is open and then closed. The value of $(A)+(B)$ is compared with the values obtained with both apertures $(A+B)$ open. If the system is linear these two values should be identical:

$$(A)+(B)=(A+B).$$

If this is not the case, the system shows nonlinearity which is proportional to the amount by which the sum of $(A)+(B)$ differs from $(A+B)$. This difference is then used to correct the transmittance values measured on the solid or liquid filters.

e. **Integrating Sphere and Photomultiplier Arrangement.** The radiations emitted from the exit slit of the monochromator and passing through the aperture or the filter are received on the target of the integrating sphere. This sphere is illustrated in figures 2, 7, 9, and 10. A block of aluminum made from identical halves was cut to produce a half sphere in each block. The halves were joined together to form a hollow sphere. Its diameter is 125 mm and a target, made from a circular plate, 35 mm in diameter, is located at the center of the sphere. The front surface of the sphere has a 20 mm diameter opening. This opening can be closed by a shutter which is operated remotely by a pneumatic system. A 50 mm diameter opening is at the opposite end to which the housing of the photomultiplier is attached by an "O" ring to provide a light-tight joint. The inside of the sphere is coated using a suspension of $BaSO_4$; the outside is painted black.

Under these circumstances the sensitive surface of the photodetector receives the radiations originating from the exit slit of the monochromator only after these radiations have undergone at least two diffuse reflections.

The photomultiplier is a 50 mm flat-faced, silica end window tube with a 44 mm cathode and 11 venetian blind dynodes having CsSb secondary emitting surfaces. The cathode is an S−20 or tri-alkali type. The spectral range of this tube is from below 200.0 nm to 850.0 nm. The operating voltage used is 850 V. The photomultiplier output is supplied to a current-to-voltage converter consisting of an operational amplifier with high precision feedback resistors with values of 10^6, 3×10^6, 10^7, 3×10^7, and $10^8\Omega$. Dark current compensation is also available. This electronic system, described in figure 13 was designed and assembled by K. W. Yee of the NBS Electronic Instrumentation Section. The output from the current-to-voltage unit is connected to a digital voltmeter, illustrated in figure 14, with one microvolt resolution on the 1 V full scale range.

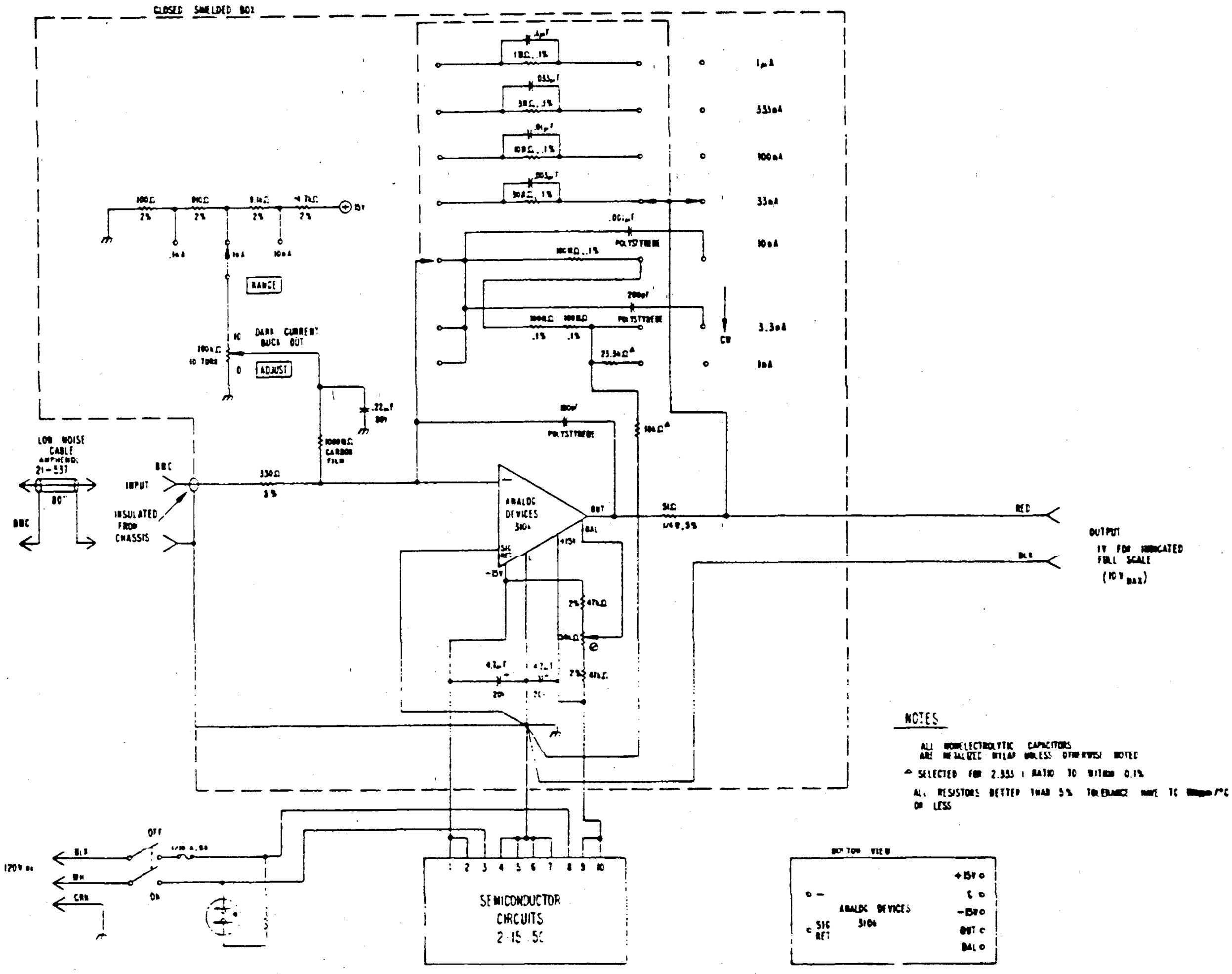


FIGURE 13. Schematic of the current-to-voltage circuitry. Courtesy of K. W. Yee.

The optical components located after the exit slit of the monochromator, including the photomultiplier tube, are enclosed in a light-tight box 200 cm long, 70 cm wide and 76 cm deep (fig. 3). The removable front panel is provided with a sliding door to permit rapid access to the filter-holder system. The box contains outlets for the compressed air which operates the apertures, sample carriage and integrating sphere shutter, and for the electrical connection from the photomultiplier. The inside walls are lined with thermal insulation painted black. When in use, all nonblack metal parts are covered with a black cloth to reduce stray light. The entire equipment is placed on a vibration isolation table 3.66 m by 1.52 m. The optical benches and the monochromator are secured by stops which are attached to the table surface. The alignment of the optical parts is made and checked periodically with a low-power laser shown in figure 3 (CW gas laser, output power 2 mW, λ 6328 Å) and with a high intensity tungsten-halogen lamp shown in figure 5.

f. **Data Collection and Presentation Systems.** The data output from the digital voltmeter (DVM), corresponding to the current generated at the photomultiplier tube by the radiations passing through the aperture system (A, B, $A+B$) or the samples (I) and blanks (I_0), can be obtained by visual means or computer operation. Both methods have been used in this work with good results. In the visual mode, the operator examines the digital voltmeter display and takes a mental average of the data. The display rate is adjusted to about one reading per second.

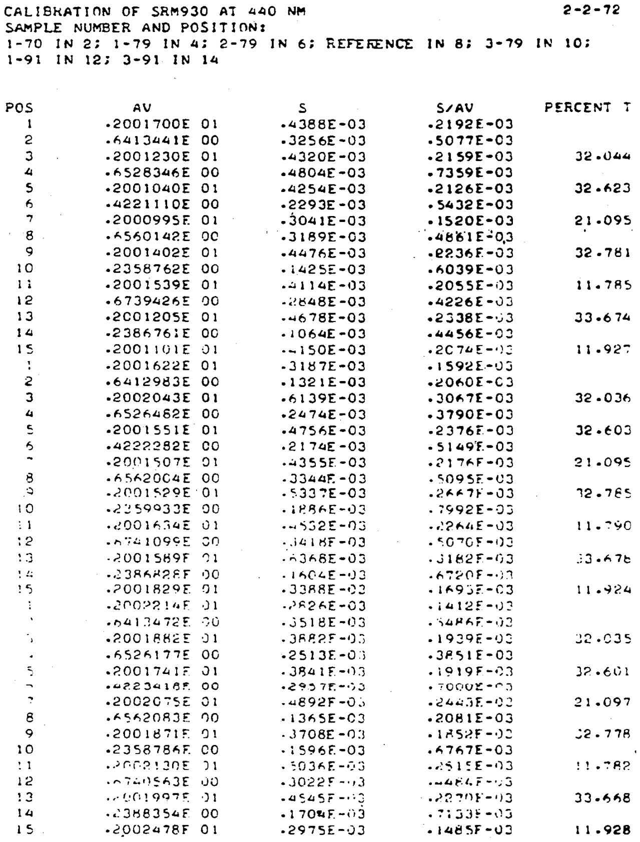
When measurements are taken by computer, the display of the digital voltmeter is adjusted to a faster rate: for instance, 10 to 20 data per second, depending on the capabilities of the instrument and measurement requirements. In our work, we use 10 data per second and collect 50 individual data for each measurement. This information is fed to the computer which calculates and prints the results as averages with the corresponding standard deviation, relative standard

FIGURE 14. Console containing the power supply for the photomultiplier tube, the dc null detector, the current-to-voltage converter, the digital voltmeter, the command panel for computer operation, and the command panel for the pneumatic operation of the shutter, aperture system, and single sample carriage. The electric switches for operating the circular neutral wedge are also located on this panel.

Middle right: potentiometer for monitoring the dc current supplied to the tungsten ribbon filament lamp.

At bottom left: teletype for data presentation.

Right: light panel which indicates the position of the automatic seven sample holder.



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CALIBRATION OF SRM930 AT 440 NM                                  2-2-72
SAMPLE NUMBER AND POSITION:
1-70 IN 2; 1-79 IN 4; 2-79 IN 6; REFERENCE IN 8; 3-79 IN 10;
1-91 IN 12; 3-91 IN 14

POS        AV              S             S/AV        PERCENT T
  1     .2001700E 01    .4388E-03    .2192E-03
  2     .6413441E 00    .3256E-03    .5077E-03
  3     .2001230E 01    .4320E-03    .2159E-03       32.044
  4     .6528346E 00    .4804E-03    .7359E-03
  5     .2001040E 01    .4254E-03    .2126E-03       32.623
  6     .4221110E 00    .2293E-03    .5432E-03
  7     .2000995E 01    .3041E-03    .1520E-03       21.095
  8     .6560142E 00    .3189E-03    .4861E-03
  9     .2001402E 01    .4476E-03    .2236E-03       32.781
 10     .2358762E 00    .1425E-03    .6039E-03
 11     .2001539E 01    .4114E-03    .2055E-03       11.785
 12     .6739426E 00    .2848E-03    .4226E-03
 13     .2001205E 01    .4678E-03    .2338E-03       33.674
 14     .2386761E 00    .1064E-03    .4456E-03
 15     .2001101E 01    .4150E-03    .2074E-03       11.927
  1     .2001622E 01    .3187E-03    .1592E-03
  2     .6412983E 00    .1321E-03    .2060E-03
  3     .2002043E 01    .6139E-03    .3067E-03       32.036
  4     .6526482E 00    .2474E-03    .3790E-03
  5     .2001551E 01    .4756E-03    .2376E-03       32.603
  6     .4222282E 00    .2174E-03    .5149E-03
  7     .2001507E 01    .4355E-03    .2176E-03       21.095
  8     .6562004E 00    .3344E-03    .5095E-03
  9     .2001529E 01    .5337E-03    .2667E-03       32.765
 10     .2359933E 00    .1886E-03    .7992E-03
 11     .2001634E 01    .4532E-03    .2264E-03       11.790
 12     .6741099E 00    .3418E-03    .5070E-03
 13     .2001589E 01    .6368E-03    .3182E-03       33.676
 14     .2386828E 00    .1604E-03    .6720E-03
 15     .2001829E 01    .3388E-03    .1693E-03       11.924
  1     .2002214E 01    .2826E-03    .1412E-03
  2     .6413472E 00    .3518E-03    .5486E-03
  3     .2001882E 01    .3882E-03    .1939E-03       32.035
  4     .6526177E 00    .2513E-03    .3851E-03
  5     .2001741E 01    .3841E-03    .1919E-03       32.601
  6     .4223416E 00    .2957E-03    .7000E-03
  7     .2002075E 01    .4892E-03    .2443E-03       21.097
  8     .6562083E 00    .1365E-03    .2081E-03
  9     .2001871E 01    .3708E-03    .1852E-03       32.778
 10     .2358786E 00    .1596E-03    .6767E-03
 11     .2002130E 01    .5036E-03    .2515E-03       11.782
 12     .6740563E 00    .3022E-03    .4484E-03
 13     .2001997E 01    .4545E-03    .2270E-03       33.668
 14     .2388354E 00    .1704E-03    .7133E-03
 15     .2002478E 01    .2975E-03    .1485E-03       11.928
```

FIGURE 15. Computer data presentation.

deviation, and sample position number to identify the measurement. When transmittance measurements are made on individual samples or when linearity checks are performed, the readings are initiated manually for every position. When the seven sample holder is used for sequential measurements, the operation is performed automatically by the computer. It is programmed to take a predetermined number of individual DVM readings (50), print the arithmetic average, followed by the standard deviation, relative standard deviation, percent transmittance and sample position (fig. 15). At the conclusion of each measurement, the computer initiates a signal which rotates the holder to the next position. This is followed by the data taking and sample changing sequence until the measurements are stopped manually or automatically by a provision made in the computer program.

The programming of the entire computer operation was developed by J. Aronson, R. Freemire, and J. Wing. The computer-instrument interfacing was performed by F. Ruegg and R. Shideler of the NBS Analytical Chemistry Division, Technical Service Group, under the supervision of J. DeVoe.

III. Stability of the Electronic System

As a rule, before taking measurements with the spectrophotometer, a warmup period of one hour is required. The room temperature is kept at 24 ± 1 °C, and the relative humidity is 35 percent. The particulate matter is controlled through special filters which rates the room in the 100,000 class.

The dark current of the photomultiplier tube was measured by taking 15 replications each consisting of the average of 50 individual digital voltmeter readings. These measurements were made using 850 V at the anode. The average dark current under these circumstances produced 0.000682 V with a relative standard deviation of 0.71 percent.

In all of our work, a dark current buck-out arrangement was used. A series of measurements were performed to determine the stability of this dark current compensation. To this effect, 15 consecutive measurements, each representing the average of 50 individual digital voltmeter readings, were made and the average dark current value was 0.000024 V with a relative standard deviation of 23.1 percent.

Four tests were made to determine the stability of the electronic system and the radiation source using the computer data acquisition mode.

a. **Stability of the Current-to-Voltage Converter.** A constant voltage was supplied to the converter using the dark current compensation provided on the unit. Fifty individual measurements were taken every 5 seconds and the average value was printed along with its percent standard deviation. The measurements were then repeated 15 times and an average of the 15 values was calculated along with the corresponding percent standard deviation. These measurements were then repeated three times. The results are summarized in table 1. This table also presents the values for the first group and the average values and corresponding percent standard deviation for the two consecutive groups. It can be seen from the stability of the current-to-voltage unit that measurements can be performed with a reproducibility of about 0.0012 (at the 67% confidence level) expressed as percent standard deviation for a single determination. The time interval between the first and last group of measurements was 15 min.

TABLE 1. Stability of the current-to-voltage converter alone measured in three groups of 15 replications each

Replication	Average of 50 individual measurements; volts	Percent standard deviation	
1	1.003494	0.0013_6	
2	1.003496	0.0012_9	
3	1.003482	0.0014_6	
4	1.003507	0.0011_1	
5	1.003515	0.0013_4	
6	1.003508	0.0013_6	
7	1.003497	0.0011_4	
8	1.003498	0.0013_3	
9	1.003505	0.0011_0	
10	1.003510	0.0014_2	
11	1.003518	0.0013_3	
12	1.003521	0.0012_0	
13	1.003522	0.0013_1	
14	1.003507	0.0012_6	
15	1.003527	0.0013_7	
Average of replications	1.003507		First group
Percent standard deviation	0.0012		
Average of replications	1.003535		Second group
Percent standard deviation	0.0015		
Average of replications	1.003545		Third group
Percent standard deviation	0.0010		

b. **Stability of the Current-to-Voltage Converter Plus the Photomultiplier Tube Supplied with 850 V and in Total Darkness.** The measurements were made as previously described and the results are presented in table 2.

c. **Stability of the Current-to-Voltage Converter and the Photomultiplier Tube Supplied with 850 V and Exposed to the Radiation of a Tritium Activated Fluorescence Source.** A constant radiation source consisting of a tritium activated phosphor was placed before the integrating sphere and a series of measurements were taken following the technique described above. Table 3 shows the results.

d. **Stability of the Current-to-Voltage Converter, the Photomultiplier Tube Supplied with 850 V, and the Tungsten Ribbon Filament Lamp.** The same measuring procedure as mentioned in a, b, and c was used here. In this case, however, the incandescent tungsten lamp was used as the source of radiation. Table 4 summarizes the results of four groups of measurements over a period of 20 min. This last series of measurements indicate that the single-beam spectrophotometer is capable of producing measurements of radiation fluxes with a percent standard deviation of about 0.022_5 for single measurements with 2.00 V at the photomultiplier tube anode.

TABLE 2. Stability of the current-to-voltage converter and the photomultiplier tube at 850 V in total darkness

Replication	Average of 50 individual measurements; volts	Percent standard deviation
1	1.012329	0.027_1
2	1.012342	0.048_8
3	1.012322	0.027_2
4	1.012320	0.033_8
5	1.012394	0.043_5
6	1.012421	0.015_3
7	1.012404	0.018_4
8	1.012406	0.029_6
9	1.012365	0.019_6
10	1.012402	0.019_1
11	1.012465	0.025_2
12	1.012412	0.061_5
13	1.012451	0.023_5
14	1.012417	0.029_6
15	1.012481	0.024_8
Average of replications	1.012395	First group
Percent standard deviation	0.0050	
Average of replications	1.012467	Second group
Percent standard deviation	0.0033	
Average of replications	1.012510	Third group
Percent standard deviation	0.0035	

In these measurements the stability of the direct current (nominal 5 V; 14 A) supplied to the tungsten ribbon lamp was monitored with the potentiometer, and the variation of this current was less than one part in 10^5 during a series of 15 consecutive measurements (5 min).

Following the four stability tests discussed earlier, a consecutive series of six measurements were made to determine the reproducibility of transmittance measurements. To this effect seven Schott NG-4 neutral glass filters were placed in the automatic sample carrying system and the data acquisition and sample changing operations were performed automatically through the computer unit. As mentioned previously, the sample carrying system can accept seven samples in positions 2; 4; 6; 8; 10; 12; 14, and eight intermediate positions 1; 3; 5; 7; 9; 11; 13; 15. The odd numbers correspond to measurements of the nonattenuated radiation beam passing through air and are marked I_0, while the even numbers correspond to measurements of the attenuated radiations after passing through the absorbing material and are marked I. The uncorrected transmittance, T, is then

$$T=\frac{I}{I_0}.$$

The radiation flux from the tungsten ribbon filament lamp was attenuated with the circular neutral wedge until a photocurrent corresponding to about 2.0020 V was obtained for the nonattenuated beam I_0. The photomultiplier tube was supplied with 850 V and the 30 MΩ resistor was used at the current-to-voltage converter. For every position, 50 digital voltmeter readings were taken by the computer at a rate of 10 to 15 per second. The average value was printed along with the sample position, the standard deviation, the relative standard deviation, and the transmittance values for the glass filters 2; 4; 6; 8; 10; 12; and 14:

$$T_2=\frac{I_2}{\frac{I_1^0+I_3^0}{2}};\quad T_4=\frac{I_4}{\frac{I_3^0+I_5^0}{2}};\quad T_6=\frac{I_6}{\frac{I_5^0+I_7^0}{2}};\quad \text{etc.}\ \ldots$$

until the seven glass filters were measured. This sequence was repeated six times and the results are given in table 5.

As can be seen from these data, the reproducibility of sequential transmittance measurements can be performed with an average standard deviation of 0.010 percent for a single determination.

IV. Wavelength Calibration

The wavelength scale of the monochromator is provided with a counter which indicates wavelength directly in ångstroms. This counter is checked for

TABLE 3. Stability of the current-to-voltage converter and the photomultiplier tube at 850 V and exposed to the radiation of a tritium activated fluorescent source

Replication	Average of 50 individual measurements; volts	Percent standard deviation
1	1.536345	0.027_5
2	1.536326	0.030_2
3	1.536196	0.022_6
4	1.536289	0.023_5
5	1.536106	0.030_9
6	1.536117	0.031_4
7	1.535916	0.025_1
8	1.536065	0.023_6
9	1.536179	0.029_3
10	1.536003	0.023_3
11	1.536083	0.021_5
12	1.535961	0.026_3
13	1.536052	0.031_2
14	1.536095	0.026_6
15	1.536092	0.026_2
Average of replications	1.536122	First group
Percent standard deviation	0.0082	
Average of replications	1.535768	Second group
Percent standard deviation	0.0095	
Average of replications	1.535522	Third group
Percent standard deviation	0.0054	

accuracy with a low pressure mercury discharge lamp placed before the entrance slit of the monochromator. The following wavelengths were used for calibration: 3650.2 Å; 4046.6 Å; 4077.8 Å; 4339.2 Å; 4347.5 Å; 4358.4 Å; 4916.0 Å; 5460.7 Å; 5769.6 Å; and 5790.7 Å. If additional reference wavelengths are needed, a Cd-Hg or a He-discharge lamp could be used for calibration. The wavelength counter was then checked using the procedure recommended by Gibson [2], and a slit of 0.1 mm which is equivalent to an effective spectral bandpass of 0.08 nm. The deviation of the wavelength counter from the true value was found to be less than ±0.1 nm; hence no wavelength correction was applied to the measurements discussed here.

V. Stray Radiation

Tests were made to determine the stray radiant energy (SRE) in the monochromator proper, as well as in the photometric arrangement. The measurement of stray radiation in the monochromator, that is, the radiation energy at wavelengths different from those of the nominal spectral bandpass transmitted through the instrument, is not easy or infallible. A detailed discussion of this instrumental parameter was given in an ASTM Tentative Method [35] and the procedure recommended in this work was used to determine SRE in the blue and yellow spectral range. In this procedure, a solution of methylene blue, which has a strong absorption in the range from λ 600 to 660 nm is used. The SRE using a slit of 1 mm (0.8 nm) was equal to or less than five parts in 10^5.

The SRE generated inside the photometric system is defined as the radiant energy which falls on the photosensitive detector without passing through the absorbing sample. This SRE is usually produced by reflections and scattering of radiations on the optical and mechanical parts located between the exit slit of the monochromator and the integrating sphere. The measurements were performed using a slit of 1 mm by placing a front surface mirror at the sample position, which reflects to the instrument all radiations received from the exit slit imaged at the mirror surface. The size of this image was about 8 mm high and 1 mm wide. In this way, a maximum SRE was generated in the spectrophotometer. The measurements were then performed at λ 577.3 nm, using a radiation flux intensity five times greater than that used in routine transmittance measurements, by determining the dark current of the photomultiplier with the shutter in the closed position at the integration sphere. An average dark current of 0.040 mV was observed. The mirror was then placed at the sample position, the shutter

TABLE 4. Stability of the current-to-voltage converter, the photomultiplier tube at 850 V, and the tungsten ribbon filament lamp

Replication	Average of 50 individual measurements; volts	Percent standard deviation	
1	2.002395	0.038_0	
2	2.001356	0.022_6	
3	2.002145	0.024_7	
4	2.000975	0.026_2	
5	2.001944	0.020_7	
6	2.000925	0.028_1	
7	2.001832	0.026_1	
8	2.000825	0.023_5	
9	2.001551	0.026_3	
10	2.000960	0.021_2	
11	2.001739	0.023_1	
12	2.000851	0.024_4	
13	2.001729	0.028_2	
14	2.000825	0.023_0	
15	2.001557	0.024_4	
Average of replications	2.001441		First group
Percent standard deviation	0.026		
Average of replications	2.001517		Second group
Percent standard deviation	0.012		
Average of replications	2.000826		Third group
Percent standard deviation	0.025		
Average of replications	2.001268		Fourth group
Percent standard deviation	0.027		

TABLE 5. Reproducibility of transmittance measurements on seven Schott NG-4 glass filters No. 2; 4; 6; 8; 10; 12; and 14

Replication No.	Percent transmittance						
	2	4	6	8	10	12	14
1	33.327	21.711	12.236	50.990	33.377	20.906	13.473
2	33.325	21.710	12.237	50.983	33.377	20.903	13.471
3	33.321	21.711	12.241	50.992	33.383	20.900	13.474
4	33.320	21.708	12.240	50.988	33.375	20.901	13.470
5	33.323	21.710	12.239	50.983	33.379	20.901	13.474
6	33.325	21.710	12.238	50.986	33.377	20.904	13.470
Average	33.32	21.710	12.238	50.987	33.378	20.902	13.472
Percent σ	0.0080	0.0051	0.0150	0.0072	0.0083	0.0108	0.0141
Average percent σ				0.010			

was opened and measurements were made again. The average value found was 0.037 mV. This indicated that no SRE could be detected under the experimental circumstances.

VI. Linearity Control

The single-beam static optical system described in this work permits the unequivocal use of the radiation addition principle by means of the double-aperture method for determining departure from linearity of the entire optical, photometric, and electronic system, and thus of the photometric accuracy of transmittance measurements.

The double-aperture and its positioning on the optical bench was described earlier. Its use will now be illustrated, and follows the procedure developed and used at the National Physical Laboratory.

Since the linearity of photometric data for a given photomultiplier tube depends on the anode voltage, the values at the current-to-voltage converter, and the ambient temperature, all measurements were made using identical experimental conditions. These same conditions were maintained when transmittance measurements were performed. Since the linearity is, within 1 part in 10^4, not usually a function of wavelength [36], all measurements were performed at λ 565.0 nm. A recent study of this parameter at NBS by Mielenz and Eckerle indicates that there may be a relation between wavelength and linearity at the level of 1 part in 10^5 [38].

The intensity of the radiation flux produced by the tungsten ribbon lamp was attenuated with the circular neutral wedge until a photocurrent equivalent to 2.0020 V was obtained when both apertures, A and B, were open. A setting of 850 V was used at the photomultiplier tube with a 30 MΩ resistor at the current-to-voltage converter. Fifty individual DVM readings were taken and the average value for $(A+B)$ was printed. Aperture B was then closed, and 50 DVM readings were taken. The average value for aperture A was printed. The average value for aperture B was then obtained in a similar manner by closing aperture A and opening aperture B. This sequence was repeated three times, ending with an $(A+B)$ value.

Identical measurements were made over a range of attenuation corresponding to 4 cascaded steps of 2 to 1 as illustrated in the actual example which follows:

Step 1	$(A+B)$	A	B
	2.0014_7	1.0159_6	0.9864_1
	2.0015_6	1.0159_9	0.9864_0
	2.0020_6	1.0160_9	0.9862_5
	2.0021_1	$1.01598 +$	$0.98635 = 2.00233$
Av.	2.00181		
Diff.	$2.00233 - 2.00181 = +0.00052$		
% Corr.	$= -0.026$		

Step 2	$(A+B)$	A	B
	1.0004_9	0.5102_0	0.4910_8
	1.0007_8	0.5099_6	0.4909_1
	1.0006_6	0.5100_4	0.4911_5
	1.0009_0	$0.51007 +$	$0.49105 = 1.00112$
Av.	1.00071		
Diff.	$1.00112 - 1.00071 = +0.00041$		
% Corr.	$= -0.041$		

Step 3	$(A+B)$	A	B
	0.5006_2	0.2565_7	0.2443_0
	0.5005_6	0.2567_3	0.2443_6
	0.5007_0	0.2566_5	0.2443_2
	0.5006_2	$0.25665 +$	$0.24433 = 0.50098$
Av.	0.50063		
Diff.	$0.50098 - 0.50063 = +0.00035$		
% Corr.	$= -0.069$		

Step 4	$(A+B)$	A	B
	0.2502_0	0.1287_2	0.1217_5
	0.2502_8	0.1285_6	0.1216_8
	0.2501_9	0.1285_4	0.1216_6
	0.2502_3	$0.12861 +$	$0.12170 = 0.25031$
Av.	0.25023		
Diff.	$0.25031 - 0.25023 = +0.00008$		
% Corr.	$= -0.031$		

The correction curve is established from these data by plotting voltages on the abscissa and the corresponding additive correction value on the ordinate. These are tabulated below and illustrated in figure 16.

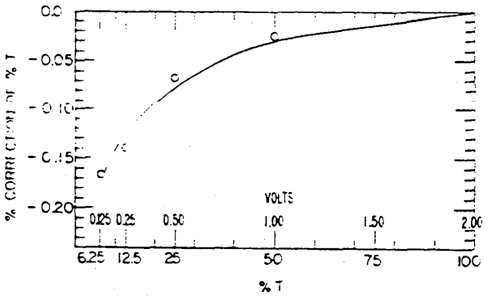


FIGURE 16. Linearity correction curve.

Voltage	% T	% Correction of % T
2.00	100	0.0
1.00	50	0.026
0.50	25	0.067
0.25	12.5	0.14
0.125	6.25	0.167

When transmittance measurements are performed, the I_0 reading is initially set with the circular neutral wedge to a value near 2.0020 V. The I value is then measured. If the initial $I_0 = 2.00214$ V and final $I_0 = 2.0022_8$ V and $I = 0.54220$ V, then percent T is:

$$\frac{0.54220}{\frac{2.00214+2.00228}{2}} \times 100 = 27.081$$

which is the noncorrected value. To correct this value, one takes from the ordinate of figure 16 the value corresponding to 0.54220 on the abscissa which, in this case, is 0.072. The corrected percent T value is then:

$$27.081 - \left(\frac{27.081 \times 0.072}{100}\right) = 27.061.$$

Mielenz and Eckerle have studied recently the double aperture method for testing photometric linearity, and have used a curve-fitting procedure for finding the nonlinearity correction rather than the method described earlier (38).

VII. Sample Position

A series of measurements were performed to determine the magnitude of error which could occur when the sample is oriented with its entrance face at an angle to the incoming radiation beam. The single sample holder provided with the rotating table, as described in section II, paragraph c, was used. Transmittance measurements were performed by producing the image of the exit slit of the monochromator at the entrance face of the sample (aperture f:10). The data are shown in table 6. The consequence of this condition on transmittance measurements is discussed by Mielenz in this Journal.

TABLE 6. Percent transmittance (%T), measured on three neutral glass filters 1.0; 1.5; and 2.0 mm thick at λ = 440 nm, at three angles of incidence

Angle of incidence	Filter 1.0 mm	Filter 1.5 mm	Filter 2.0 mm
Normal incidence	32.91_5	19.83_8	11.60_6
1°	32.89_9	19.83_3	11.60_4
2°	32.89_7	19.81_9	11.59_9
3°	32.88_7	19.81_2	11.58_8

Similar measurements were made to determine the identity of positions on the seven-sample automatic changer described in section II, paragraph c. For this experiment, seven neutral glass filters A; B; C; D; E; F; and G were used and were positioned in holders 2; 4; 6; 8; 10; 12; and 14 in three different arrangements as described by Garfinkel, Mann and Youden [39].

TABLE 7. Evaluation of the identity of the seven stations of the automatic sample changer

Percent Transmittance (%T) at station number (St. No.) 2; 4; 6; 8; 10; 12; and 14 for filters A, B, C, D, E, F, and G at λ = 465.0 nm

Run No.		A	B	C	D	E	F	G
I	%T	27.08	16.44	46.39	26.12	15.22	37.34	23.23
	St. No.	2	4	6	8	10	12	14
II	%T	27.07	16.40	46.39	26.11	15.21	37.34	23.23
	St. No.	14	2	4	6	8	10	12
III	%T	27.08	16.43	46.39	26.11	15.21	37.33	23.23
	St. No.	10	12	14	2	4	6	8

Transmittance measurements were then performed on all filters for the three different arrangements and the results are given in table 7. From these data it can be concluded that the seven stations are interchangeable and will produce measurements which will not differ by more than one part in one thousand.

VIII. Influence of Polarized Radiations on Transmittance Measurements

This effect was determined by measuring the transmittance of a Schott NG–4 neutral glass filter at four wavelengths using radiations emerging from the predisperser-monochromator unit, and by projecting the image of the exit slit (8 by 0.5 mm) at the entrance face of the filter with a convergent beam geometry corresponding to an f:10 opening. The glass filter was checked prior to measurements with a polariscope for freedom of internal tensions. Column one of table 8 shows the results obtained when transmittance measurements were made using the radiations produced by the spectrophotometer. Column two shows the results obtained when a polarizing sheet, with the vibration plane horizontal, was placed in front of the glass filter. The measurements obtained with the vibration plane in vertical position, are given in column three.

These measurements show that polarized radiations can affect transmittance measurements of solid glass filters when noncollimated beam geometry is used. This effect is predicted by the Fresnel equations mentioned in the introduction and should disappear when collimated radiations are used (1, pg. 100).

XI. Comparison of Transmittance Measurements

Two sets of solid filters were used in a comparative test to determine the reproducibility of transmittance measurements between two laboratories. One set was made from three neutral glass Schott NG–4 filters having nominal percent transmittances of 10; 20; and 30. The second set was made as described elsewhere [4]. Three evaporated metal (Inconel) on fused quartz (nonfluorescent) plates having nominal percent

TABLE 8. Effect of polarization on percent transmittance (%T) measured at four wavelengths on a Schott NG–4 glass filter

Wavelength nm	%T Produced by spectro-photometer 1	%T Polarizer, plane of vibration horizontal 2	%T Polarizer, plane of vibration vertical 3	% Difference %T_1 – %T_3
440.0	19.81_8	19.80_0	19.78_8	– 0.15
465.0	22.59_7	22.60_0	22.56_9	– 0.12
590.0	19.17_8	19.17_0	19.09_9	– 0.41
635.0	20.61_1	20.60_2	20.54_7	– 0.31

transmittances of 25; 50; and 75 were used. The transmittance measurements were performed on two sets of filters at the National Physical Laboratory (NPL) in England using their high accuracy spectrophotometer, and at NBS on the instrument described in this paper. The measurements at NBS were carried out before and after the measurements at NPL. All measurements were made with noncollimated convergent beam geometry. A rectangular surface of the filter about 3 mm by 8 mm was used at NPL and the beam was only slightly convergent. At NBS an area about 8 mm by 0.5 mm was used for the transmittance measurements.

The results given in table 9 indicate that an average difference of –0.19 percent of the values was obtained between the measurements carried out at NPL and at NBS. An average difference of –0.30 percent of the value was found when similar measurements were performed on the inconel-on-quartz filters, as shown in table 10.

X. Standard Reference Materials for Spectrophotometry

The need for providing means and materials to check the proper functioning of a spectrophotometer was discussed in some detail in previous publications [3, 4]. At that time it was established that the accuracy of the photometric scale is a critical and most demanding parameter in spectrophotometry. Hence, particular attention was given to a number of ways for checking this parameter. Investigations showed that solid colored glass filters, exhibiting optical neutrality over the spectral range from 400.0 nm to 700.0 nm, would constitute an acceptable Standard Reference Material (SRM). From the various colored glass filters available, Schott NG–4 "neutral glass" was selected, prepared and characterized. It is now offered by NBS as a means to check the photometric scale of spectrophotometers.

TABLE 9. Comparison between the percent transmittances (%T) measured on three Schott NG–4 glass filters at NPL and NBS

Wavelength nm	NBS, %T March 12, 1971	NBS, %T May 18, 1971	NBS, %T average	NPL, %T February 1971	% Diff. NBS to NPL
440.0	12.92	12.91	12.91_5	12.93	– 0.11
465.0	14.96_5	14.98	14.97_3	15.01	– 0.25
590.0	11.70	11.64	11.67	11.67	0.0
635.0	12.72	12.68	12.70	12.72	– 0.16
440.0	19.62_5	19.58	19.60_3	19.62	– 0.09
465.0	22.38_5	22.35	22.36_7	22.43	– 0.28
590.0	19.06	18.95	19.00_5	19.01	– 0.03
635.0	20.45_5	20.37	20.41_3	20.47	– 0.23
440.0	32.89	32.86	32.87_5	32.98	– 0.32
465.0	35.52	35.54	35.53	35.66	– 0.36
590.0	31.16_5	31.10	31.13_5	31.21	– 0.25
635.0	32.56_5	32.52	32.54_3	32.62	– 0.24

Average difference between NBS and NPL percent T values = – 0.19 percent.

Table 10. Comparison between the percent transmittances (%T) measured on three inconel-on-silica filters at NPL and NBS

Wavelength nm	NBS, %T 1	NBS, %T 2	NBS, %T average	NPL, %T	% Diff. NBS to NPL
450.0	24.87	24.88	24.87_5	24.93	− 0.18
550.0	23.78	23.82	23.80	23.86	− 0.25
650.0	23.38	23.39	23.38_5	23.46	− 0.32
450.0	49.35	49.33	49.34	49.56	− 0.44*
550.0	47.60	47.60	47.60	47.81	− 0.44
650.0	46.85	46.85	46.85	47.14	− 0.64
450.0	72.17	72.20	72.18_5	72.30	− 0.16
550.0	72.05	72.11	72.08	72.20	− 0.17
650.0	72.20	72.34	72.27	72.33	− 0.08

Average difference between NBS and NPL percent T values = − 0.30 percent.

*This filter had a flaw in the form of a crack which was sometimes visible and other times invisible. The larger differences found in the measurements of this filter may be due to this flaw.

SRM 930, developed in the Analytical Chemistry Division and available since March 1971 consists of three glass filters. Each filter bears an identification number, and the upper left corner has been removed to indicate correct orientation in the metal holder (fig. 17).

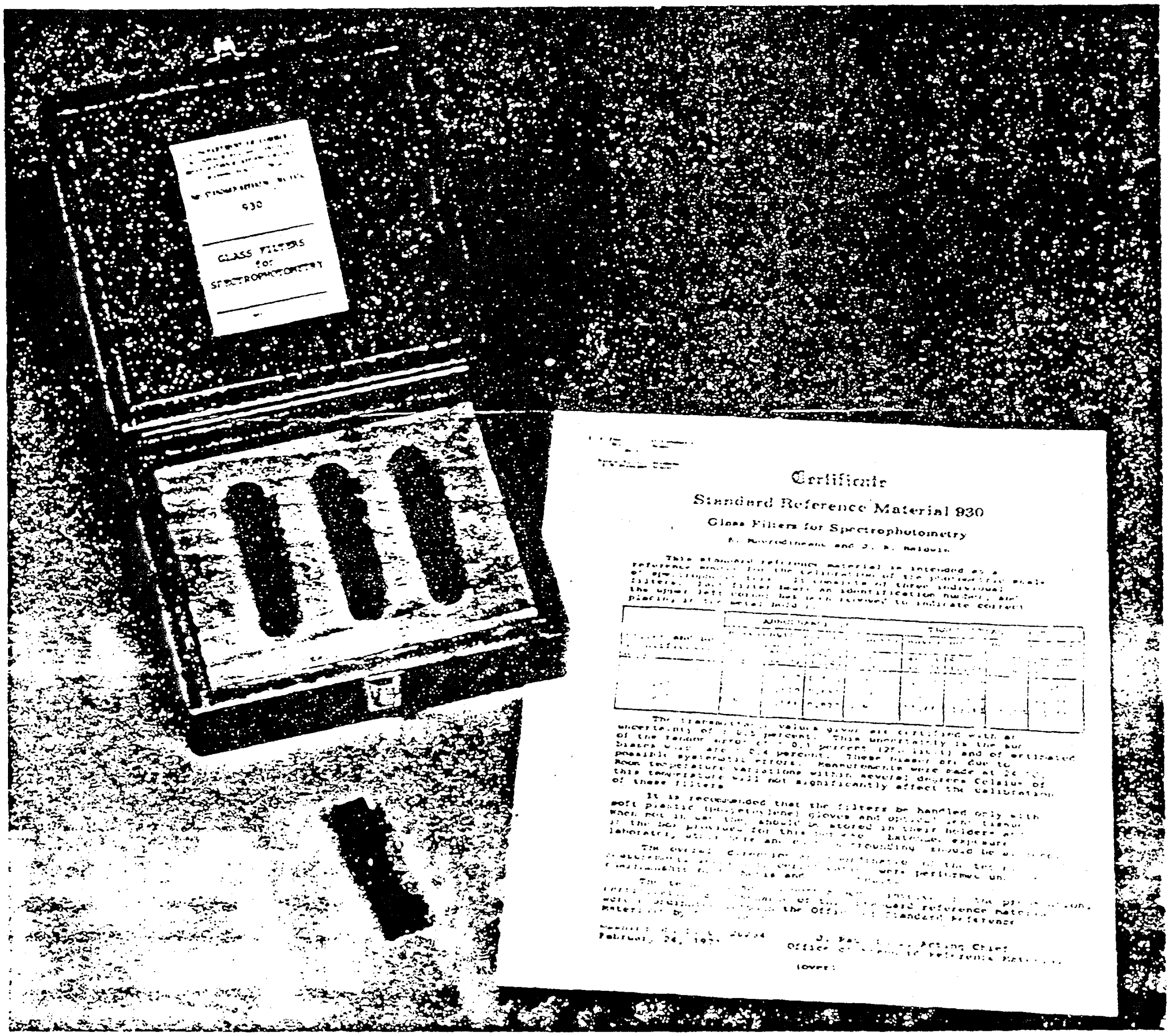
The transmittance measurements were made with the high accuracy spectrophotometer described in this paper, and are certified with an uncertainty of ±0.5 percent of the value. This uncertainty is the sum of the random errors of ±0.1 percent (2SD limit) and of estimated biases which are ±0.4 percent. These biases are due to possible systematic errors originating principally from the inherent inhomogeneity and instability of the glass as well as from positioning of the filter. Measurements were made at 24 °C, and variations within several degrees Celsius of this temperature will not significantly affect the calibration of the filters. The neutral NG−4 glass for the filters was provided by Schott of Mainz, Germany and is designated as "Jena Colored and Filter Glass." Nominal transmittance for a filter 1.5 mm thick is 20 percent at 400.0 nm wavelength and 32 percent at 700.0 nm wavelength. Between these limits the transmittance varies in a monotonic manner.

The filter is held in a frame and the size and shape of the filters and frames were selected, for practical considerations, to conform to the dimensions of the standardized cuvettes for which holders are supplied in most conventional spectrophotometers. The filters are approximately 1.0, 1.5, and 2.0 mm thick. Corresponding to these thicknesses are nominal transmittances of 30, 20, and 10 percent, respectively. These thicknesses were selected to provide a means for calibrating the photometric scale at three different levels.

The effective spectral bandpasses used to determine the certified values were equal to or smaller than 2.2 nm at 440.0 nm; 2.7 nm at 465.0 nm; 5.4 nm at 590.0 nm; and 6.0 nm at 635.0 nm. The transmittance measurements are made by producing the image of the slit (about 8 mm by 0.5 mm) using a convergent beam geometry with an opening of f:10 corresponding to an angle of 7° to 8° in the middle of the entrance face of the filter. This beam geometry was used to reproduce the average experimental conditions found in most of the conventional spectrophotometers available today. Prior to the certification, each filter is examined for surface defects and thoroughly cleaned. If, through handling, the surface of the filter becomes contaminated, it may be cleaned with a small soft brush attached to a rubber tube connected to a vacuum source [40]. If contamination results from fingerprints, they must be removed before making measurements. This may be accomplished by removing the filter from its holder, breathing lightly on it, and rubbing the surface gently with optical lens tissue. The clean filter is then properly positioned in its holder. To remove and replace the filter in the holder, the spring-loaded plate should be lifted with care to prevent damage to the filter. As little handling as possible is recommended. SRM 930 should be used according to the directions on the certificate; consult the manufacturer of the instrument if differences are obtained that exceed those specified by the manufacturer.

Under no circumstances should other cleaning procedures which make use of detergent solutions, organic solvents, etc. be applied.

When a filter has become contaminated beyond cleaning by the procedure described in the certificate, it should be forwarded to NBS. After proper cleaning, the filters will be checked and, if needed,



Certificate

Standard Reference Material 930

Glass Filters for Spectrophotometry

FIGURE 17. NBS Standard Reference Material 930 and calibration certificate.

recalibrated using the high accuracy spectrophotometer described in this work.

It was already stated that the accuracy of photometric scales defines only one of the parameters required for obtaining accurate transmittance values and molar absorptivities. Other factors must also be established. These are wavelength accuracy, adequate spectral bandpass, stray light, cell parameters (when solutions are measured), fluorescence, polarization, reflection, and temperature coefficient. Some of these variables were discussed in NBS Technical Notes 544 and 584 and are also examined in this paper.

The transmittance data given in the certificate which accompanies each SRM 930 depend not only on the intrinsic properties of the glass and the experimental measurement conditions, but on the surface state of the glass. This parameter varies with time and exposure conditions. When glass is exposed to normal room atmosphere and temperature, its surface is corroded to an extent depending on the composition, time of exposure, concentration, temperature and nature of the glass surface acting agents. This action produces a change in the reflecting and transmitting properties of the material [41]. For instance, a well-known phenomenon called "blooming" of the glass is due to the formation of an SiO layer at the surface of the glass. This layer, which increases the transmittance, acts as an antireflection coating. The speed with which such a layer is formed varies with the composition of the glass, the atmosphere and time. Generally speaking, several years are needed for a fresh surface to reach equilibrium. This, and similar phenomena are presently being studied, along with means to stabilize the surface state of glass filters. Until more information is acquired in this field, we recommend that the colored glass filters issued as SRM 930 be rechecked annually to determine whether any physicochemical changes, which might affect the transmittance values, have occurred.

Another important factor is the need for defining and producing a clean glass surface. Until now the final cleaning of the NG–4 filters was made with redistilled ethyl alcohol and pure water (thermally distilled and deionized). Other cleaning procedures are under consideration. The use of isopropyl alcohol in vapor or liquid form associated with mild ultrasonic action is being investigated [41].

The transmittance characteristics of the SRM 930 limit the use of this material to the visible region of the spectrum from about 400 nm to the near infrared. Since the ultraviolet region, from about 200 nm is also important to the analyst who uses spectrophotometric methods, exploratory work is underway to select and certify solid material for checking the photometric scale in this spectral region. Optical filters exhibiting small transmittance-wavelength dependence in the spectral range 200 nm to near infrared can be obtained by evaporating thin semitransparent layer of a metal on a suitable transparent substrate [42, 4], and such filters have been considered in this work. The metals selected were inconel and chromium which exhibit adequate transmission characteristics and good adhesion to the substrate. The substrate was nonfluorescent fused quartz. A series of filters were prepared by the optical shop at NBS according to the following specifications: a number of nonfluorescent optical quality fused quartz plates, 10 mm × 25 mm and 1 mm thick, were cut and polished, Inconel or chromium metal was evaporated on the surface to produce nominal transmittances of 25, 50, and 75 percent. The surface bearing the evaporated metal was coated with a layer of optical cement which was transparent to the visible and ultraviolet radiations down to 230 nm. A clear plate of the same material was used to cover and protect the evaporated metal layer.

The filter assembly was then marked at one corner to insure its proper positioning and the finished filter was placed in a metal holder of conventional size (approximate o.d. 13 x 13 x 57 mm) fitting the cuvette holder found in most spectrophotometers. The metal holder was also marked at one side to permit positioning of the filter in a reproducible manner.

In addition to the evaporated metal filters, a number of units were prepared using only the clear uncoated fused quartz plates and assembled with the same optical cement. When desired, these clear filter assemblies could be used as reference samples in the blank compartment.

Before submitting the evaporated metal filters to transmittance measurements, a study was made of the effect of radiations on their transmittances. A filter was exposed to an accelerated test in which radiations had the same spectral distribution as the fluorescent lighting of the laboratory, except that they were 1000 times more intense. The filter was exposed for an equivalent of 36,000 hours of continuous irradiation. This test was made on a radiation accelerator made available by the Building Research Division of NBS. The percent transmittance was measured before and after the exposure and gave the following results:

	Transmittance, percent			
Wavelength, nm	250	380	500	650
Before exposure	44.48	51.35	48.90	47.41
After exposure	44.11	51.34	48.92	47.47

The relative standard deviation for a single determination of these measurements was 0.01 percent. As can be seen, the only significant relative change in transmittance of about 0.84 percent of the value occurred at 250 nm.

Several sets of these filters were calibrated at five selected wavelengths, 250 nm; 350 nm; 450 nm; 550 nm; and 650 nm, using the cleaning and measuring procedures outlined for Schott NG–4 colored glass neutral filters. The results indicated that the reproducibility of transmittance measurements is good (percent standard deviation 0.009 to 0.024) and is comparable to those obtained for the colored glass filters at all wavelengths except 250 nm. From the experimental data, it is evident that the transmittance of the evaporated metal filter at 250 nm is critical and, at present, no satisfactory explanation for this phenomenon can be given. A limitation of the evaporated metal filters is that they attenuate the intensity of radiation by *reflecting* a part of it, rather than absorbing. This can produce, in certain circumstances, undesirable stray light in the instrument and make the transmission measurements dependent on the geometry of the optical beam. However, since these filters are closer to optical neutrality than the colored glass filters, and since they can be used in the ultraviolet region as well, they were included in this work.

This limitation was apparent from the data obtained in a cooperative study conducted at C. Zeiss by A. Reule using conventional spectrophotometers. On the other hand, a similar comparative test, made on the same filters by F. J. J. Clarke at NPL has produced the results presented in table 10. One can observe that, in spite of the limitations mentioned above, an agreement within −0.30 percent of the value was obtained between NBS and NPL measurements at the indicated wavelength.

Further studies will be needed to assess unambiguously the transmittance characteristics of evaporated metal-on-quartz filters, with or without a protective quartz plate, and to assess their suitability as Standard Reference Materials to check the photometric scale of spectrophotometers in the ultraviolet and visible part of the spectrum.

XI. Addendum

The identification of commercial instruments and products, is given in the Addendum only to permit reproduction of the work described in this paper. In no instances does such identification imply recommendation or endorsement by the National Bureau of Standards, nor does it imply that the particular equipment or product is necessarily the best available for the purpose.

Radiation source for visible—Microscope lamps, type 18A/T10/1P-6V: General Electric Co., Lamp Division, Nela Park, Cleveland, Ohio 44112. For ultraviolet: Atlas single coil halogen (Bromine) lamp, type P1/8, 30V, 250W: GTE Sylvania, Inc., 6610 Electronic Drive, Springfield, Virginia 22151.

Power supply for microscope lamp, Kepco, Model JQE 15-50-M-VP: Kepco, Inc., 131-38 Sanford Avenue, Flushing, New York 11352. For tungsten-halogen single filament lamp: same manufacturer, Model JQE-36-30 Mt-VP.

Potentiometer: Leeds and Northrup Model K3 with null meter and power supply. Resistors: Leeds and Northrup 0.1, 50 A and 0.01, 100 A: Leeds and Northrup, Sumneytown Pike, North Wales, Pa. 19454.

Nonfluorescent fused silica: Dynasil Corporation of America, Berlin, New Jersey 08009.

Neutral Density Attenuator and $BaSO_4$ white paint: Eastman Kodak Co., Special Products Sales, Kodak Apparatus Division, Elmgrove Plant, Rochester, New York 14650.

Monochromator with predisperser: McPherson Instrument Corp., 530 Main Street, Acton, Massachusetts 01720.

Optical benches with carriers and $x-y$ sample holder with micrometer control: Gaertner Scientific Corp., 1201 Wrightwood Ave., Chicago, Illinois 60614.

Lens holders: Ardel Instrument Co., Inc., P. O. Box 992, Jamaica, New York 11431.

Ball bushing and rails: Thompson Industries, Inc., Manhasset, New York 11030.

Pneumatic cylinders and accessories: Clippard Instrument Laboratory, Inc., Cincinnati, Ohio 45239.

Rotating table: Ealing Optics Division, 2225 Massachusetts Avenue, Cambridge, Massachusetts 02140.

Thermostating holders for glass cells and glass filters: Cary Instruments, 2724 South Peck Road, Monrovia, California 91016.

Pneumatic ratchet system: Allenair Corp., P. O. Box 350, 255 East 2nd Street, Mineola, New York 11501.

Black paint—Nextel 101–c 10 Black: Reflective Products Division 3M, 2501 Hudson Road, St. Paul, Minnesota 55101.

Photomultiplier tube EMI–9558QA: Gencom Division, 80 Express Street, Plainview, New York 11803.

Power supply for photomultiplier tube: Model 415B and digital voltmeter 8400A: John Fluke Manufacturing Co., P.O. 7428, Seattle, Washington 98133.

Thermal insulation: Photoshroud, Shumway Optical Instruments Corp., 2118 Beechgrove Place, Utica, New York 13501.

Vibration isolation table: Lansing Research Co., 705 Willow Avenue, Ithaca, New York 14850.

Low power laser: Model 195 cw gas laser, output power 2 mw: Optics Technology, Inc., 901 California Avenue, Palo Alto, California 94304.

Tritium activated fluorescent source, Beta light Marker HM–110: Canrad Precision Industries, Inc., 630 Fifth Avenue, New York, New York 10020.

Colored glass neutral filters, Schott NG–4: Fish-Schurmann Corp., 70 Portman Road, New Rochelle, New York 10802.

Spectral lamp: Oriel Optics Corp., 1 Market Street, Stamford, Connecticut 06902.

Polarization filters: Polaroid Corp., 119 Windsor Street, Cambridge, Massachusetts 02139.

Computer: 24K memory and 16 bit words, EMR computer, Division of Weston Instruments, Inc., Schlumberger Co., 8001 Bloomington Freeway, Minneapolis, Minnesota 55420.

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J. I. Shultz, Assistant Section Chief, Analytical Coordination Chemistry (NBS), for critically reviewing the manuscript.

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Liquid Absorbance Standards

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Errors in the measurement of the absorbances of liquid filters result from instrumental and chemical uncertainties. This paper presents a systematic study of these variables on the absorbances of selected filters. Three types of liquid filters are discussed. These are (1) individual solutions of high purity compounds, (2) composite mixtures and (3) aqueous solutions of organic dyes. The accuracy of the absorptivity data is established using NBS-calibrated glass filters. The magnitude of the errors arising from spectral bandpass, beam geometry, stray light, internal multiple reflections, and refractive index are delineated. Finally, as a practical outgrowth of this study, the development and issuance of NBS Standard Reference Material 931, Liquid Absorbance Standards for Ultraviolet and Visible Spectrophotometry, is described.

Key words: Absorptivity data; accuracy; liquid absorbance standards; Standard Reference Materials.

I. Introduction

In the use of filters for checking the accuracy of the photometric scale of spectrophotometers, one needs materials which exhibit absolute spectral neutrality. Such ideal filters are not available at present. The use of the materials which have been suggested is limited by instrumental and sample variables. For this reason, one must carefully specify conditions which define the other sources of errors.

The first part of this paper reviews several of the more important instrumental parameters and discusses their effects on absorbance measurements. Examples from the literature are given to illustrate the magnitude of the errors arising from spectral bandpass, stray light, nonparallel radiation, and multiple reflections. The second part presents some experimental studies of several materials which may serve as calibration "filters" or standards.

II. Instrumental Parameters

A. Wavelength Accuracy

The absorbances of most samples are sufficiently wavelength dependent that even in the most favorable regions of maxima and minima a wavelength error of 1−2 nm can produce absorbance errors of a few tenths of a percent. When performing measurements on the slopes of absorption peaks, the wavelength setting is obviously much more critical and errors are typically several percent per nanometer. Wavelength accuracy, therefore, becomes especially important when making absorbance measurements at, for example, an isosbestic point.

A line source provides the most definitive means for establishing the accuracy of the wavelength scale. The best single source is a mercury lamp which may be used throughout the range of 200–1000 nm. For calibration in the visible region, helium lines are also useful.

Gibson [1][1] has discussed the use of mercury and helium sources and has listed those lines best suited for wavelength calibration. Other sources which have been used include neon, cadmium, cesium, and sodium. The M.I.T. Wavelength Tables [2] summarizes the most prominent emission lines of these elements. Not all wavelengths are given and care must be taken in using any of these lines in order not to confuse closely adjacent lines with the one being checked. For those spectrophotometers having a hydrogen or deuterium source, the emission lines at 486.13 and 656.28 nm (H) or 485.99 and 656.10 nm (D) may provide convenient checks at these wavelengths.

Calibrated holmium oxide and didymium glasses may be useful secondary standards, particularly for checking recording spectrophotometers in which a dynamic check of the instrument is often desirable. The apparent absorption maxima of these filters, however, may vary with spectral bandpass. Therefore, for the highest accuracy, they should be calibrated for the spectral bandpass at which they are to be used.

In general, prism instruments require more frequent calibration than the grating type because their dispersion is temperature dependent. Corrections in the visible region are of the order of 0.1 nm per degree Celsius. Hysteresis effects must also be considered

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[1] Figures in brackets indicate the literature references at the end of this paper.

and the wavelength settings should always be approached from the same direction.

B. Finite Bandwidth

Spectral bandpass differences undoubtedly account for a significant portion of the discrepancies in molar absorptivity values reported in the literature. Some have resulted from the employment of inadequate instrumentation; others have resulted from improper use of these instruments or complications arising from solution equilibria.

Increasing the spectral bandpass at an absorption maximum has two effects on apparent peak heights: (1) the observed values are always less than the true values and (2) the differences between the two are proportionally greater at higher absorbances. Thus, for a finite bandpass, a plot of absorbance versus concentration or path length will always have a smaller slope than it does in monochromatic radiation and, in addition, will be concave to the concentration axis. Figure 1 illustrates these effects. The reverse behavior will be observed for measurements at an absorption minimum. Meehan [3] has given a simple example which verifies these two effects mathematically.

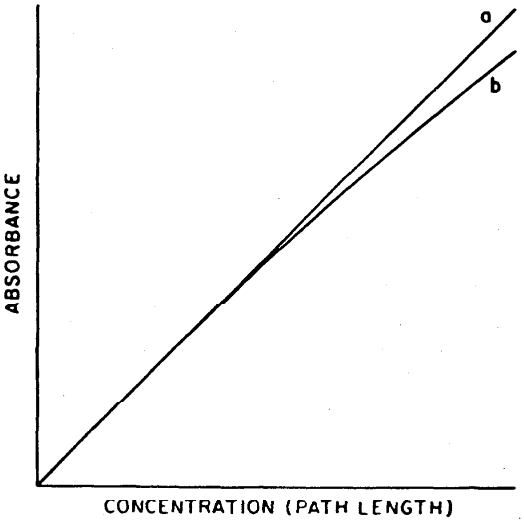


FIGURE 1. Effect of spectral bandpass on absorbance: (a) Monochromatic radiation (b) Finite bandwidth.

Consider that an absorbing system is illuminated by two monochromatic radiations of wavelengths λ_1 and λ_2 and that the Lambert-Beer law is obeyed at each wavelength with absorptivities a_1 and a_2. If the effective intensities of the two wavelengths are I_1 and I_2, respectively, the transmittance, T, is

$$T = (I_1 \cdot 10^{-a_1bc} + I_2 \cdot 10^{-a_2bc})/(I_1 + I_2) \tag{1}$$

where b = path length and c = concentration. If λ_1 is the wavelength of maximum absorption so that $a_1 > a_2$, then

$$T = \frac{1}{(1+r)} \cdot 10^{-a_1bc}[1 + r \cdot 10^{(a_1-a_2)bc}] \tag{2}$$

where $r = I_2/I_1$. The absorbance, A, defined as $-\log T$ is

$$A = a_1bc + \log(1+r) - \log[1 + r \cdot 10^{(a_1-a_2)bc}] \tag{3}$$

which may be differentiated with respect to bc to give

$$dA/d(bc) = a_1 - [r(a_1 - a_2)10^{(a_1-a_2)bc}]/[1 + r \cdot 10^{(a_1-a_2)bc}] \tag{4}$$

At low absorbances

$$dA/d(bc)_{bc \to 0} = \frac{a_1 + ra_2}{1+r} \tag{5}$$

whereas when bc becomes large, $10^{a_1bc} \gg 10^{a_2bc}$ and the limiting slope is

$$dA/d(bc)_{bc \to \infty} = a_2. \tag{6}$$

Thus, the limiting absorptivity at low absorbances is a weighted average of a_1 and a_2 while, at higher absorbances, it is equal to a_2, the smaller of the individual absorptivities.

The theory for the correction of spectral bandpass errors has been developed by Hardy and Young [4], Eberhardt [5], and Broderson [6]. Rigorous calculation requires integration of the relation

$$A_{\text{obs}} = \log \frac{\int I_\lambda S_\lambda d\lambda}{\int I_\lambda S_\lambda 10^{-A} d\lambda} \tag{7}$$

where I_λ is the incident intensity and S_λ is the spectral sensitivity of the detector. Equation (7) emphasizes the fact that the observed absorbance depends not only on the shape of the absorption curve, but also on the wavelength distribution of the source and the detector response. If $I_\lambda S_\lambda$ is constant over the wavelength interval used, approximate corrections for spectral bandpass may be calculated by assuming a Gaussian shape for the absorption peak and a triangular slit function for the instrument. Some calculations are given in table 1 based on these assumptions. The tabulated values of A_{obs}/A agree closely with those observed experimentally for A up to 1. As shown by Broderson [6] however, for values of RBW larger than 0.5, A_{obs}/A also depends on A and the above treatment is no longer applicable.

C. Stray Light

Stray light is defined as any light outside the spectral region isolated by the monochromator that reaches the detector. It is produced by scatter from the optics and

TABLE 1. Dependence of A_{obs} on spectral bandpass at an absorption maximum under idealized conditions (see text)

RBW[a]	A_{obs}/A	RBW[a]	A_{obs}/A
0.0100	0.99995	0.0800	0.9970
.0200	.9998	.0900	.9962
.0300	.9995	.1000	.9954
.0400	.9992	.2000	.9819
.0500	.9988	.3000	.9604
.0600	.9983	.4000	.9321
.0700	.9977	.5000	.8987

[a] Relative bandwidth: Ratio of spectral bandwidth to half bandwidth of fully resolved peak.

walls of the monochromator and is present in varying amounts in all spectrophotometers.

Stray light can lead to varied problems in spectrophotometry. Spurious absorption bands may arise in some cases. More frequently, however, deviations from the Lambert-Beer law are produced. These deviations are positive if the stray light is absorbed and negative if it is not.

Extensive literature exists on the detection, measurement and mimization of stray light. Several of the more useful papers are those by Hogness, Zscheile and Sidwell [7], Perry [8], Slavin [9], and Poulson [10] in which additional references may be found.

The amount of stray light present is proportionally large in those wavelength regions where the transmission of the monochromator, the source intensity or the detector sensitivity are relatively low. These regions, which should first be checked, are 200–230 nm and 600–700 nm. If a tungsten lamp must be used in the range 350–400 nm, a visible cutoff filter should always be employed.

The quantitative assessment of stray light requires the use of extremely sharp cutoff filters. Slavin [9] and Poulson [10] have described a number of liquid and solid filters that may be used. Aqueous solutions of the alkali halides, for example, are extremely useful in the ultraviolet. Care must be taken, however, that light leaks in the cuvet compartment are not contributing to the observed results. The apparent stray light will not vary appreciably with slitwidth if only instrumental stray light is present. In a quick test for light leaks, the apparent stray light will decrease rapidly as the slits are opened.

The stray light error encountered most frequently in spectrophotometry produces a decrease in an absorption peak and leads to negative deviations from the Lambert-Beer law. If it is assumed that none of the stray light is absorbed by the sample, the measured absorbance is related to the true transmittance by the expression [9]

$$A_{\text{obs}} = \log\,[(1-T)\,S+T]^{-1} \qquad (8)$$

where S is the instrumental stray light expressed as a fraction. The effects of several levels of unabsorbed stray light on absorbance are given in table 2. Most samples absorb sufficiently in other regions of the spectrum to filter out a proportion of the stray light. Thus, indirect estimates of instrumental stray light based on deviations from linearity are generally low.

TABLE 2. Effect of unabsorbed stray light on absorbance

A	A_{obs}		
	$S=0.0001$	$S=0.001$	$S=0.01$
0.1	0.1000	0.0999	0.0989
.5	.4999	.4990	.4907
1.0	.9996	.9961	.9626
1.5	1.499	1.487	1.384
2.0	1.996	1.959	1.701

D. Nonparallel Incident Radiation

A perfectly parallel beam of radiation must come from a point source and can carry only an infinitesimal amount of energy. The light beam in a spectrophotometer always has some finite angular size. As a result, the average light path is greater than the perpendicular distance between the cuvet faces. Meehan [3] has considered the case in which the incident radiation is convergent or divergent in one dimension such as corresponds, respectively, to radiation focused by a cylindrical lens or leaving a narrow slit. If Θ and R are the angles of incidence and refraction, respectively, the path length of the extreme ray (fig. 2) is $b/\cos R$ and the absorbance of this ray is $A/\cos R$. Other rays enter at smaller angles and the observed absorbance is given by

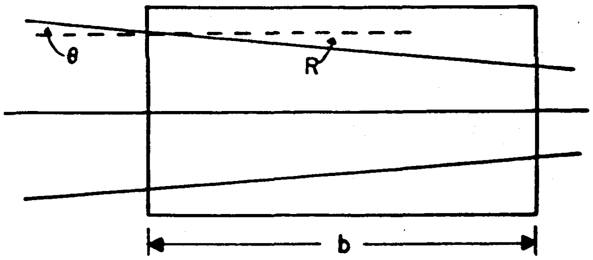


FIGURE 2. Nonparallel incident radiation [3]. Reprinted by permission of John Wiley & Sons, Incorporated.

$$A_{\text{obs}} = A/R_{\max}\int_0^{R_{\max}} 1/\cos R\,dR. \qquad (9)$$

For this relation to hold, the incident beam must be uniform over its cross section. Evaluation of this integral for $R_{\max}$ of 2–10° ($\Theta_{\max}=2.7°–13.4°$) gives the following percent errors in A (table 3). Thus, for angles of incidence up to 5 degrees, the absorbance error is less than 0.1 percent.

E. Multiple Reflections

On passage of light through a cuvet containing solvent or sample solution, some radiation is reflected

TABLE 3. Dependence of absorbance error on nonparallelism of incident radiation

$R_{\max}$, degrees	$\Theta_{\max}$, degrees	Percent error in A
2	2.7	0.020
4	5.3	.081
6	8.0	.18
8	10.7	.33
10	13.4	.51

at each of the two air-glass and the two glass-liquid surfaces. For perpendicular incidence, the fraction f reflected on passing from a medium of refractive index n_1 to a second having refractive index n_2 is given by the Fresnel expression

$$f=\left[\frac{n_1-n_2}{n_1+n_2}\right]^2 \tag{10}$$

For an air-glass surface, $f=0.04$ and for a glass-liquid surface, $f=0.0035$. When more than one surface is involved, the effects of multiple reflections must be considered. The essential question when dealing with liquid filters is whether the solvent completely compensates for such reflections. According to Goldring et al. [11], it does not. They consider the case where the reflections from all surfaces perpendicular to the light beam on the two sides of the sample are grouped together to form two effective reflection coefficients, r_1 and r_2, as shown in figure 3. Considering only first and second order reflections, the observed absorbance is

$$A_{\text{obs}}=\log\frac{P_1}{P_2}=\log\frac{P_0}{P_0T}\,\frac{(1-r_1+r_1r_2)}{(1-r_1+r_1r_2T^2)} \tag{11}$$

which, after transformation and series expansion, can be reduced to the following expression:

$$A_{\text{obs}}=A+0.4343\,(1-T^2)\,\frac{r_1r_2}{1-r_1}. \tag{12}$$

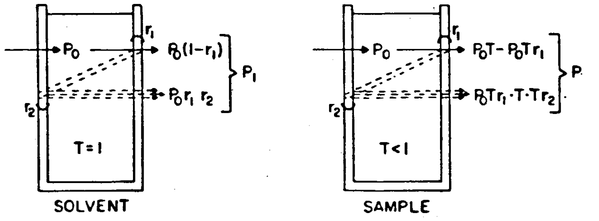


FIGURE 3. Internal multiple reflections [11]. Reprinted by permission of the American Chemical Society.

Considering only the reflections from the two cuvet faces and that $r_1=r_2=0.05$, the variation of A_{obs} with A is

A	0.1	0.2	0.5	1.0	2.0
A_{obs}	0.1004	0.2007	0.5010	1.0011	2.0011

Thus, because of internal multiple reflections, the measured absorbance should always be larger than the true absorbance with the percentage difference being greatest at low absorbances. Verification of these deviations is difficult experimentally. Goldring et al. [11], however, have suggested ways of minimizing reflection effects. These include positioning the cuvet at a small angle to the beam and stopping down the detector surface, immersing the cuvet in a fluid contained in a larger rectangular cuvet turned at an angle to the beam, or by using a cuvet with prismatic windows. In each case, the effective path length must be determined separately. Other alternatives consist of using cuvets constructed of absorbing materials or immersion of a glass filter into the sample. For a limited wavelength range, reflection errors can also be reduced by coating the external window surfaces with anti-reflection layers.

In addition to reflections from the cuvets, reflections from other surfaces must also be considered. Gibson [1] has pointed out that reflections may occur from the surfaces of the slit and detector and has suggested a means for checking this source of error. A thin glass plate is placed in its normal position at right angles to the beam and then at a small angle from this position. In the second position the reflected energy is directed out of the beam while scarcely affecting the true absorbance.

F. Cuvets

The majority of absorbance measurements are performed on solutions and, in such instances, the cuvet becomes an integral part of the measurement system. Presently, the uncertainty in the length of the light path can be a limiting factor in the determination of molar absorptivity. Nonparallelism of the end windows can be of even greater consequence. However, the effect can be minimized by using the same cuvet orientation for all measurements. The uncertainty in path length is nevertheless greater.

Cuvets are available in a variety of shapes and sizes and may be made of glass or silica. Construction remains largely an art and cuvets may vary considerably in their quality. Two methods of assembly are commonly used. These are: (1) fusion using only heat and (2) the use of intermediate, low-melting glasses. At present, each has its disadvantage. The first technique is the more desirable, but our experience, primarily with rectangular cuvets, indicates that the edges are not always completely fused. These surfaces exhibit capillarity and may become serious sources of contamination. Under such conditions, the cleaning of these cuvets is difficult, if not impossible. The use of low-melting fluxes on the other hand can produce strains because of differences in coefficients of expansion and cuvets so constructed are generally more fragile.

Cuvets are frequently offered in matched sets. This may be considered more of a convenience than a necessity since this terminology is normally used only to describe the transmission of the window material.

Path length and parallelism of end plates are of more fundamental importance. Unfortunately, no generally accepted tolerances have been established in this country for the construction of cuvets. The British Standards Institution, however, has published a set of specifications [12] which merit our consideration and possible adoption. They recommend that the path length be specified to ± 0.02 mm and the end windows be flat over a defined beam area to four Newton fringes per centimeter in mercury green light. It should be feasible for greater accuracy, especially for the 10 mm cuvets, to calibrate a limited number by appropriate metrology techniques to ± 0.01 mm.

III. Evaluation of Selected Liquid Filters

This section is a report of our efforts to develop well characterized liquid absorbance standards. Three general types of liquid filters are discussed. These are (1) individual solutions of high purity materials, (2) empirical mixtures and (3) aqueous solutions of organic dyes. Much of the emphasis has been placed on determining the optimum conditions for preparation of these filters and, once established, what the absorbing species or ionic compositions of these systems are. By combining this information with the instrumental considerations developed in the first part of this paper, it is hoped that the resulting data represent a step toward obtaining more meaningful absorptivity measurements.

A. Instrumentation

Absorbance measurements at a fixed wavelength were performed manually on a high precision double-beam spectrophotometer provided with a double monochromator. The accuracy of the photometric scale of this instrument was established with the NBS high-accuracy spectrophotometer described by Mavrodineanu [13]. The wavelength scale was checked with a mercury lamp. Potassium halide solutions [9, 10] were used to assess stray light in the ultraviolet.

Room temperature was maintained between 24 to 26 °C. The cuvet compartment and jacketed holders were thermostated by a circulating water bath. Copper-constantan thermocouples were used to measure temerature differences between the bath and the sample solution.

An electronic feeler gauge [13] was used to measure the path length of the 10 mm rectangular cuvets normally used. Solutions, assumed to obey Beer's law, were used for calibrating the 1 mm cuvets.

The accuracy of the microbalance was established with NBS-calibrated weights. Class A, 1-liter volumetric flasks were checked gravimetically. All dilutions were subsequently made by volume. To minimize errors from cell positioning, borosilicate, Pasteur-type pipets were used to transfer solutions to and from the cuvets.

B. Individual Solutions of High Purity Compounds

1. Potassium Dichromate. Numerous attempts have been made to use chormium (VI) solutions as ultraviolet absorbance standards. In the early studies [7, 14, 15, 16, 17] alkaline solutions of potassium chromate were preferred. More recently, potassium dichromate in slightly acidic media has been described [18, 19, 20]. The absorption spectra of these two systems are quite different with the latter giving the more desirable arrangement of maxima and minima (fig. 4).

The approximate composition of chromium (VI) solutions may be derived from the following equilibria:

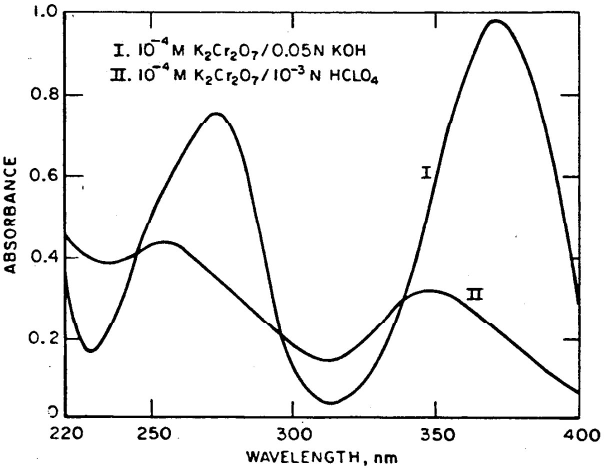


FIGURE 4. Absorption spectra of 10^{-4} M Cr (VI) in 0.05 N KOH and 10^{-3}N $HClO_4$.

$$H_2CrO_4 \overset{K_1}{=} H^+ + HCrO_4^- \tag{13}$$

$$HCrO_4^- \overset{K_2}{=} H^+ + CrO_4^= \tag{14}$$

$$2HCrO_4^- \overset{K_3}{=} Cr_2O_7^= + H_2O. \tag{15}$$

Some values reported for the equilibrium constants K_1, K_2, and K_3 at 25 °C are as follows:

$K_1 = 0.16$ [21]

$K_2 = 3.2 \times 10^{-7}$ [22] and 3.0×10^{-7} [23]

$K_3 = 43.7$ [21], 35.5 [24] and 33.0 [25].

Thus, at a pH greater than 10, chromium (VI) exists almost entirely as $CrO_4^=$ ions while, in weakly acidic solutions, the predominant species is $HCrO_4^-$ which partially dimerizes to $Cr_2O_7^=$. The amount of dimer will increase with increasing chromium concentration. The formation of H_2CrO_4 or $CrO_4^=$ can be essentially eliminated by maintaining the acidity near pH 3.

Much of the uncertainty that has arisen from the use of acidic potassium dichromate solutions as potential absorbance standards can be attributed to the dimerization reaction (eq (15)). It is primarily this equilibrium which leads to the observed deviations from Beer's law. Because of the range of values reported for K_3, one of the first objectives of the present study was to redetermine this constant. The spectrophotometric method was chosen and is described in some detail below.

a. **Spectrophotometric Determination of the Dimerization Constant for: $2HCrO_4^- = Cr_2O_7^= + H_2O$.** If $HCrO_4^-$ and $Cr_2O_7^=$ are the only Cr(VI) species present with molar absorptivities ϵ_1 and ϵ_2, respectively, the apparent molar absorptivity, ϵ_m, will be given by

$$\epsilon_m = (1-\alpha)\epsilon_1 + 1/2 \cdot \alpha\epsilon_2 \tag{16}$$

where α is the fraction of total chromium in the dichromate form. From eq (15) the thermodynamic equilibrium constant of the dimerization reaction is

$$K_3 = \frac{[Cr_2O_7^=]}{[HCrO_4^-]^2} \cdot \frac{\gamma_2}{\gamma_1^2} \tag{17}$$

or, in terms of α

$$K_3 = \frac{\alpha}{2(1-\alpha)^2 Cr_T} \cdot \frac{\gamma_2}{\gamma_1^2} \tag{18}$$

where γ_1 and γ_2 are the activity coefficients of $HCrO_4^-$ and $Cr_2O_7^=$, respectively, and Cr_T is the total chromium concentration. Because the ionic strength never exceeded 0.01, the activity coefficients may be treated by the Debye-Hückel expression [25]

$$-\log \gamma_i = \frac{AZ_i^2 I^{1/2}}{1+I^{1/2}} \tag{19}$$

where Z is the ionic charge, I is the ionic strength and A has a value of 0.509 l/mol at 25 °C. Equations (16) and (18) can be rewritten to give, respectively,

$$\epsilon_m = \alpha(1/2\,\epsilon_2 - \epsilon_1) + \epsilon_1 \tag{20}$$

and

$$\log K_3 = \log \frac{\alpha}{2(1-\alpha)^2 Cr_T} - \frac{2AI^{1/2}}{1+I^{1/2}}. \tag{21}$$

Assuming a value of K_3, one can calculate from eq (21) the α values at various total chromium concentrations. If the choice of K_3 is correct, the measured values of ϵ_m should lie on a straight line of slope $(1/2\,\epsilon_2 - \epsilon_1)$ and intercept ϵ_1, as seen from eq (20). The best value of K_3 is then determined by the method of least squares.

1. Experimental Procedure

The chromium (VI) solutions used in the subsequent studies were prepared from NBS SRM–136c, potassium dichromate, which had been dried at 110 °C for two hours. The distilled water was shown to be free of reducing impurities by titration with a dilute potassium permanganate solution. Perchloric acid was used to maintain the pH at 3.0 ± 0.1.

The optimum wavelengths for the determination of K_3 were chosen as follows: Two solutions were prepared, one containing 391 mg $K_2Cr_2O_7$/l and the other, 40.0 mg $K_2Cr_2O_7$/l. The former was transferred to a 1.023 mm cuvet and the latter to a 10.00 mm cuvet. A differential scan was then made on a recording spectrophotometer equipped with a 0–0.1 absorbance (full scale) slidewire, using the less concentrated solution as the reference. The resulting spectrum is shown in figure 5. Relatively large differences in absorbance are seen in the wavelength regions near 275 and 385 nm even though the total number of chromium atoms in each light path is the same. Thus, it is in these two regions that the largest deviations from Beer's law should be observed. Three wavelengths were subsequently selected in each region for the determination of K_3–390, 385 and 380 and 280, 275 and 270 nm, respectively.

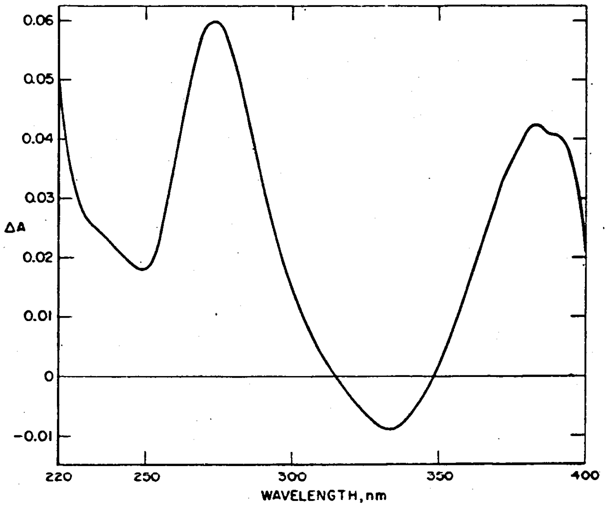


FIGURE 5. Difference spectrum of 391 mg $K_2Cr_2O_7$/l in 1.023 mm cuvet versus 40.0 mg $K_2Cr_2O_7$/l in 10.00 mm cuvet; pH = 3.

2. Results

The apparent molar absorptivities calculated from absorbance measurements on the four chromium concentrations used for the determination of K_3 are given in table 4. Using the method of least squares, these ϵ_m values were plotted against the α fractions calculated from eq (21) for various assumed values of K_3. The molar absorptivities were weighted according to the reproducibility of the absorbance measurements. A computer was used for all calculations.

A typical graph of the residual standard deviation of the experimental ϵ_m values from the best straight line fit is shown in figure 6 for K_3 values of 28 through 36. The minimum in this curve is the best estimate of K_3 and, for the case shown, is 32.6. The K_3 values at the six wavelengths are summarized in table 5.

TABLE 4. Apparent molar absorptivities, ϵ_m, of $K_2Cr_2O_7$ solutions at 25 °C; pH = 2.9 ($HClO_4$)

$K_2Cr_2O_7$ Conc. mg/l	ϵ_m l/mol^{-1} cm^{-1}					
	390 nm	385 nm	380 nm	280 nm	275 nm	270 nm
100.06	589.2	748.7	865.1	1579.3	1718.6	1881.0
400.48	714.1	878.6	997.8	1758.2	1914.4	2069.8
699.86	806.2	973.8	1094.0	1890.0	2053.5	2208.0
1000.27	875.9	1045.6	1166.5	1988.2	2161.5	2313.6

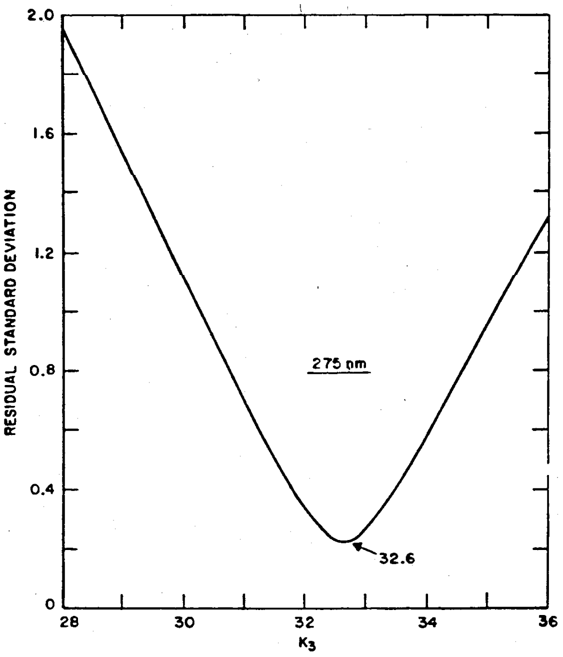


FIGURE 6. Least squares determination of K_3.

TABLE 5. K_3 values at 25 °C as determined at several wavelengths

λ, nm	K_3
390	32.4
385	33.3
380	33.8
280	33.5
275	32.6
270	31.6
Avg.	32.9
Std. Dev.	0.8
Rel. Std. Dev.	2.5%

b. Absorption Spectra of $HCrO_4^-$ and $Cr_2O_7^=$. The absorbances of three solutions containing nominally 20, 40, and 60 mg $K_2Cr_2O_7$/l (pH = 3.0 ± 0.1) were measured in a 10.00 mm cuvet at 5 nm increments over the wavelength range of 220–400 nm. A close approximation of $\epsilon(HCrO_4^-)$ was obtained by extrapolating the apparent molar absorptivities to zero chromium concentration. This extrapolation is simplified since the variation of α is nearly linear over this concentration range. Similar absorbance measurements were performed on three solutions containing 1000 mg $K_2Cr_2O_7$/l using a 1.023 mm cuvet and the corresponding α value was calculated from eq (21), with $K_3 = 32.9$. The first approximations of $1/2 \cdot \epsilon(Cr_2O_7^=)$ values were obtained by substituting the extrapolated $\epsilon(HCrO_4^-)$ results and the above α value into eq (20). The $\epsilon(HCrO_4^-)$ and $1/2 \cdot \epsilon(Cr_2O_7^=)$ values were then refined by successive approximations. The calculated absorption spectra of these two ions are shown in figure 7.

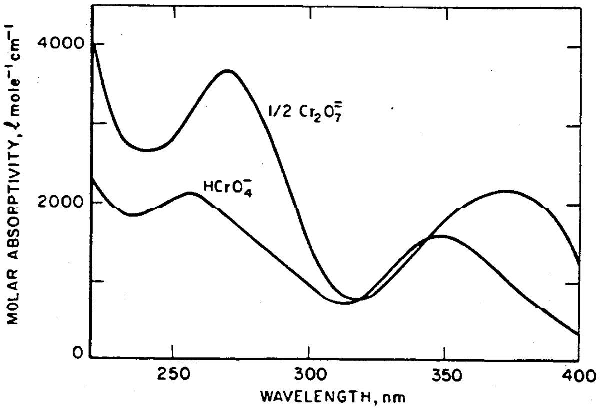


FIGURE 7. Absorption spectra of $HCrO_4^-$ and $Cr_2O_7^=$.

c. Molar Absorptivities of $HCrO_4^-$ and $Cr_2O_7^=$ at 350, 313, 257, and 235 nm. As shown previously in figure 4, the absorption spectrum of weakly acidic solutions of $K_2Cr_2O_7$ exhibit maxima at 350 and 257 nm and minima at 313 and 235 nm. These maxima and minima do not shift significantly over the concentration range of 20–100 mg $K_2Cr_2O_7$/l. This is considered a practical range for most applications since it yields absorbances of about 0.1–1.5 for a 10 mm cuvet.

The molar absorptivities of $HCrO_4^-$ and $Cr_2O_7^=$ at 350, 313, 257, and 235 nm were determined by the procedure described above and are given in table 6. The $\epsilon(HCrO_4^-)$ values are estimated to be accurate to ± 0.5 percent. Because of the imprecision of the K_3 determination, the uncertainty of the dichromate molar absorptivity values may be as large as ± 5 percent.

TABLE 6. Molar absorptivities, ϵ, of $HCrO_4^-$ and $Cr_2O_7^=$ at 25 °C

	ϵ, l mol^{-1} cm^{-1}			
	350 nm	313 nm	257 nm	235 nm
$HCrO_4^-$	1576	711	2100	1804
$1/2 \cdot Cr_2O_7^=$	1788	823	3156	2688

Table 7 shows the agreement between the experimental and calculated apparent molar absorptivities for five concentrations of potassium dichromate in dilute perchloric acid ($pH = 2.92 \pm 0.02$). The experimental values are the averages obtained on five individual samples measured at each concentration. The 95 percent confidence limits for any given set of measurements was 0.1 percent. The α values used are also given. These are seen to vary nearly linearly over the range of chromium concentrations employed.

d. Optimum pH and Choice of Acid. The chromium (VI) solutions used in this study were prepared in dilute perchloric acid media having a pH of about 3. This acidity was selected because the existing equilibria data indicated this pH limited the formation of either H_2CrO_4 or $CrO_4^=$ to less than 0.1 percent of the total Cr (VI) concentration. Perchloric acid was chosen instead of sulfuric to prevent the formation of chromate-sulfate complexes. Tong and King [24] and Davies and Prue [25] noted slight differences in the absorbances of dilute perchloric and sulfuric acid solutions after correcting for differences in ionic strength. They attributed these differences to mixed complex formation. More recently, Haight et al. [26] have shown that the conversion of $HCrO_4^-$ to $CrSO_7^=$ is quantitative in 1 *M* HSO_4^- solutions and that absorption spectrum of this complex is significantly different from the $HCrO_4^-$ spectrum.

Because most of the previous work reported in literature has been done in sulfuric acid, it was considered of interest to intercompare these two systems. The results are shown in each acid between pH 2 and 3. At 350 nm the results obtained in perchloric acid are consistently 0.1–0.2 percent higher than in sulfuric; at 235 nm, they are lower. The relatively large increase in absorptivity at 235 nm at pH 1.90, together with the above considerations, suggest the formation of additional chromium (VI) species in systems employing dilute sulfuric acid solutions. (See table 8.)

e. Effect of Temperature. The absorbances of a solution containing 60 mg $K_2Cr_2O_7$/l in dilute perchloric acid (pH = 3) were measured over the temperature range of 17 to 37 °C. The percentage change in absorbance for wavelengths of 350, 313, 257, and 235 nm is shown in figure 8. The results are uncorrected for

TABLE 7. Experimental and calculated values (E, C) of the apparent molar absorptivity of $K_2Cr_2O_7$ at 25 °C; $pH = 2.9(HClO_4)$

$K_2Cr_2O_7$ Conc. mg/l	α		ϵ_m, l mol^{-1} cm^{-1}			
			350 nm	313 nm	257 nm	235 nm
20.22	0.0090	E	1577.6	713.5	2113.6	1814.7
		C	1577.9	711.9	2108.7	1812.1
		Diff.	−0.3	+1.6	+4.9	+2.6
40.09	0.0178	E	1579.9	713.3	2120.8	1820.2
		C	1579.8	713.0	2118.8	1819.7
		Diff.	+0.1	+0.3	+2.0	+0.5
60.12	0.0262	E	1581.4	713.8	2127.2	1827.1
		C	1581.5	713.8	2126.7	1827.2
		Diff.	−0.1	0.0	+0.5	−0.1
80.17	0.0344	E	1584.0	715.0	2137.6	1835.8
		C	1583.3	714.8	2135.3	1834.4
		Diff.	+0.7	+0.2	+2.3	+1.4
99.92	0.0423	E	1585.5	715.6	2144.0	1841.5
		C	1584.0	715.0	2143.7	1841.4
		Diff.	+1.5	+0.6	+0.3	+0.1

TABLE 8. Apparent absorptivities, *a*, of sulfuric and perchloric acid solutions of $K_2Cr_2O_7$ at 25°C; $K_2Cr_2O_7$ conc. = 0.050 g/l

		a, l g^{-1} cm^{-1}			
	pH	350 nm	313 nm	257 nm	235 nm
H_2SO_4	1.90	10.71_0	4.83_7	14.44_4	12.43_7
	1.98	10.71_2	4.83_9	14.44_0	12.41_8
	2.20	10.72_2	4.83_0	14.42_9	12.40_2
	3.00	10.73_3	4.84_4	14.42_7	12.38_1
$HClO_4$	1.99	10.72_7	4.84_8	14.44_8	12.39_6
	3.08	10.74_0	4.84_5	14.43_4	12.38_3

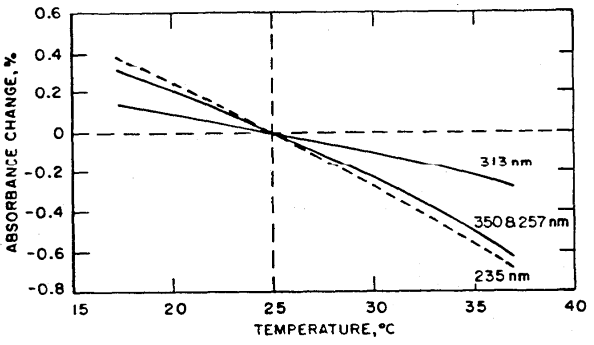


FIGURE 8. Change in absorbance with temperature for acidic solutions of $K_2Cr_2O_7$; pH = 3 ($HClO_4$).

thermal expansion of the solvent. The absorption minimum at 313 nm exhibits the smallest temperature dependence (−0.02 percent per °C) while the maxima at 350 and 257 nm and the minimum at 235 nm have temperature coefficients of −0.05 percent per °C over the range 20 to 30 °C.

f. $K_2Cr_2O_7$ in 0.5 M Na_2HPO_4 and 0.05 M KOH. To overcome some of the difficulties in handling strongly alkaline chromate solutions, Johnson [27] has recommended dissolving $K_2Cr_2O_7$ in 0.05 M Na_2HPO_4 at pH 9. The apparent molar absorptivities obtained in this medium and in 0.05 M KOH are given in table 9. As seen from the data, both systems appear to exhibit deviations from Beer's law of about three parts per thousand over the concentration range studied. Although this deviation is the same magnitude and direction predicted from uncompensated internal multiple reflections discussed previously, we have not been able to demonstrate experimentally that such reflections are responsible for the observed results. Studies in which the cuvet windows are coated with various antireflection layers are continuing however.

TABLE 9. Apparent molar absorptivity of potassium chromate in 0.05 *M* Na_2HPO_4 and 0.05 *M* KOH at 25 °C

K_2CrO_4 Conc., $M \times 10^5$	pH	ϵ_m, l mol^{-1} cm^{-1}	
		373 nm	274 nm
0.05 *M* Na_2HPO_4			
7	9.2	482_7	370_3
14	9.1	482_0	369_7
21	9.1	481_3	369_2
0.05 *M* KOH			
7	....	483_0	370_5
14	....	482_4	369_8
21	....	481_4	369_1

From a consideration of the equilibrium data, the absorbances in the two systems should be nearly identical. The ionic strength of the 0.05 *M* Na_2HPO_4 is 0.15 and, from Neuss and Rieman's work [22], the ionization constant for the reaction $HCrO_4^- = H^+ + CrO_4^=$ is approximately 9×10^{-7} mol/liter. Thus, at pH 9.1, only about 1 in 1000 chromium atoms is present as $HCrO_4^-$. Since this species is less absorbing than $CrO_4^=$ at 373 and 274 nm, the apparent molar absorptivities in 0.05 *M* Na_2HPO_4 should agree with those obtained in 0.05 *M* KOH to within 0.1 percent.

The absorbance of alkaline chromate solutions decreases with increasing temperature at both absorption peaks. Between 17 and 37 °C, the temperature coefficient at 373 nm is −0.09 percent per °C; at 274 nm, it is −0.06 per °C.

2. Potassium Nitrate. Aqueous solutions of potassium nitrate exhibit a nearly symmetrical absorption peak with λ_{max} at 302 nm. This system has been studied extensively by Vandenbelt [14] and by the British Photoelectric Spectrometry Group in two collaborative tests [28, 29]. Molar absorptivities reported vary from 7.06 to 7.20 liter mol^{-1} cm^{-1}. This range of values undoubtedly results in part from the fact that KNO_3 solutions deviate significantly from Beer's law.

Careful reexamination of this system has yielded the following apparent molar absorptivities (table 10). These are seen to decrease rather markedly with increasing potassium nitrate concentration. Several factors conceivably contribute to this behavior. In addition to possible multiple reflection effects already discussed some ionic interactions are undoubtedly possible in this system because of the relatively high concentrations of solute required coupled with the fact that the N−O bond in the nitrate ion has some polar character.

The potassium nitrate used in the above study was analyzed for commonly encountered impurities. After drying at 110 °C, the water content was less than 0.02 percent. Flame emission analyses [31] indicated the following levels of alkali and alkaline earth impurities (in parts-per-million): Li (< 0.2); Rb (13); Mg (0.2); Ca (1); Sr(4) and Ba (< 2).

At 302 nm, the absorbance of KNO_3 solutions decreases with increasing temperature. Over the range 17 to 37 °C, the temperature coefficient is −0.14 percent per °C.

Solutions stored for up to six months showed no measurable change (≤ 0.1 percent) in absorbance. Edisbury [29] has cautioned, however, that some solutions may develop bacterial whiskers on standing. Sterilization may be effected by prior boiling of the distilled water.

TABLE 10. Apparent molar absorptivity, ϵ_m, of aqueous solutions of potassium nitrate at 25 °C

	ϵ_m, l mol^{-1} cm^{-1}
KNO_3 Conc., *M*	302 nm[a]
0.028	7.16_0
.056	7.14_2
.084	7.12_7
.112	7.10_6
.140	7.09_1

[a] Spectral bandpass = 1.0 nm

3. Potassium Hydrogen Phthalate. High purity potassium hydrogen phthalate (KHP) is readily available. It is presently issued by the National Bureau of Standards as a primary acidimetric standard. Its use in the preparation of buffer solutions of known pH is well established. It has also been used as a spectral standard in the comparative evaluation of spectrophotometers [11, 32].

The absorbance of KHP solutions is dependent on acidity, as shown in figure 9. In the present study, dilute perchloric acid solutions were employed to minimize changes in ionic composition as a function of pH. Acid concentrations much above 0.1 *N* could not be used because of the precipitation of potassium perchlorate. Subsequently, all solutions were prepared to contain 10 ml of perchloric acid per liter with a pH of 1.3.

Ringbom [21] has given the first ionization constant of phthalic acid as 1.6×10^{-3} at 25 °C at an ionic strength of 0.1. At the pH used, the absorbing system consisted of 97 percent phthalic acid and 3 percent hydrogen phthalate ions. Table 11 summarizes the absorptivity values obtained on these solutions.

Absorbance decreases at the 275.5 nm maximum and increases at the 262 nm minimum with increasing temperature. Over the range 17 to 37 °C the temperature coefficients are −0.05 percent and +0.05 percent per °C, respectively.

Perchloric acid solutions of KHP (NBS-SRM 84g) were examined for possible fluorescence, using a high sensitivity spectrofluorometer. With 280 nm excitation, a faint fluorescence was detected with λ_{max} at approximately 350 nm. The possible effects of this fluorescence on the absorbance data remain to be established.

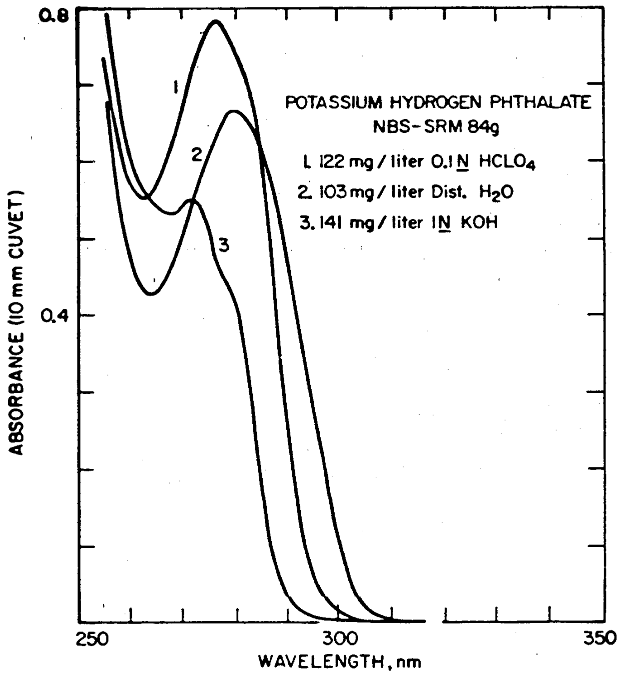


FIGURE 9. Absorption spectra of KHP solutions at different acidities.

TABLE 11. Apparent absorptivity, a, of potassium hydrogen phthalate in dilute perchloric acid at 25 °C; 10 ml $HClO_4$/liter

NBS SRM 84g Conc., g/l	a, l g^{-1} cm^{-1}	
	275.5 nm	262 nm
0.034	6.33_2	4.49_4
.142	6.31_9	4.48_9

4. Cobalt and Nickel Sulfates and Perchlorates. Aquo cobalt (II) and nickel (II) ions have relative broad absorption peaks with λ_{max} at 512 and 394 nm, respectively (fig. 10). Solutions of either may serve as useful absorbance standards. Most of the attention to date, however, has been directed at cobalt sulfate which was first recommended by Gibson [1].

In this study, the apparent molar absorptivities of $Co(H_2O)_6^{+2}$ and $Ni(H_2O)_6^{+2}$ were examined in sulfate and perchlorate media. All solutions were prepared from the high purity metals by dissolution in H_2SO_4–HNO_3 or $HClO_4$–HNO_3 acids. Nitrate was removed by repeated fuming until the acidity was reduced so that on dilution to volume, a pH of 1 could be obtained. The results obtained in the two acid media are given in table 12. The values in sulfate solution are significantly higher than in perchlorate media for both cobalt and nickel, suggesting that some complexation of these ions by $SO_4^=$ or HSO_4^- has occurred. A differ-

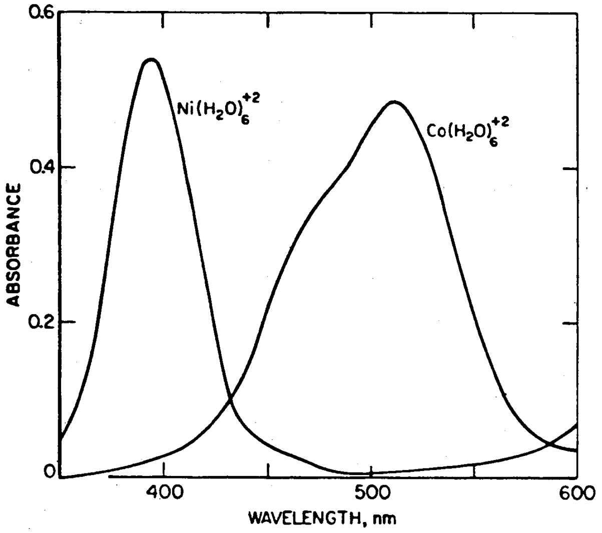


FIGURE 10. Absorption spectra of $Ni(H_2O)_6^{+2}$ and $Co(H_2O)_6^{+2}$.

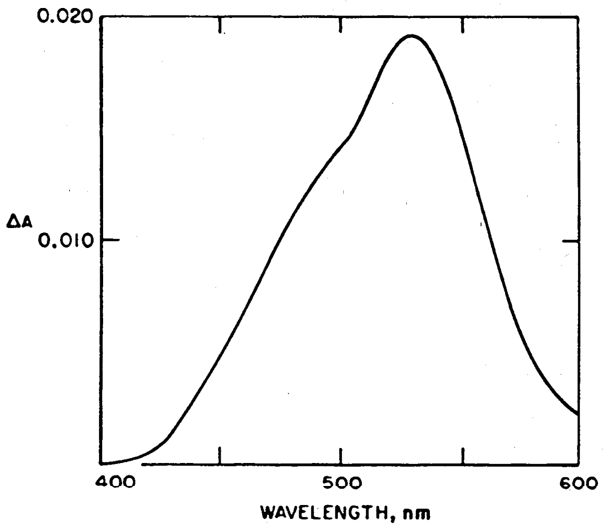


FIGURE 11. Differential spectrum of cobalt sulfate versus cobalt perchlorate; 10 mm cuvets; pH = 1.

TABLE 12. Apparent molar absorptivities, ϵ_m of acidic solutions of cobalt and nickel ions at 25 °C; pH = 1.0

Co Conc. g/l	ϵ_m (512 nm) l mol^{-1} cm^{-1} Sulfate	ϵ_m (512 nm) l mol^{-1} cm^{-1} Perchlorate
2.4	4.88_1	4.80_3
12.0	4.86_9	4.79_1

Ni Conc. g/l	ϵ_m (394 nm) l mol^{-1} cm^{-1} Sulfate	ϵ_m (394 nm) l mol^{-1} cm^{-1} Perchlorate
2.3	5.16_7	5.09_0
11.5	5.15_4	5.07_7

ential spectrum of 0.2000 molar cobalt solution in sulfate against the same cobalt concentration in perchlorate media is shown in figure 11. In sulfate solution, the formation of a second cobalt species with $\lambda_{max} = 528$ nm is indicated. Similar behavior was also observed for nickel.

The absorbances of cobalt and nickel solutions at their maxima are temperature dependent and increase with increasing temperature. Between 17 and 37 °C the temperature coefficients are 0.18 and 0.14 percent per °C at 512 and 394 nm, respectively. To explain their nuclear magnetic resonance data on cobalt(II) solutions, Swift and Connick [33] have suggested that the octahedral $Co(H_2O)_6^{+2}$ complex is in equilibrium with the tetrahedral $Co(H_2O)_4^{+2}$ complex and that formation of the latter is favored by increasing temperature. By analogy with halide complexes, the tetrahedral configuration would be expected to be approximately one hundred times more absorbing than the octahedral complex. A very small shift in this equilibrium could result in a relatively large change in absorbance and thereby account for the large temperature coefficient of this system. The same explanation may also be valid for nickel solutions.

C. Liquid Filters of Improved Optical Neutrality

Two approaches have been used in an attempt to improve the optical neutrality and extend the wavelength range of liquid filters. One is based on the use of empirical mixtures, while the other employs water-soluble, organic "black" dyes.

In 1946, Thomson [34] described the preparation of a grey inorganic solution consisting of chromic, cobaltous and cupric sulfates and potassium dichromate. This solution, however, does not transmit below 300 nm. To extend the wavelength range farther into the ultraviolet, a second exploratory solution (henceforth referred to as the NBS composite) was prepared in which the cupric and potassium dichromate components of the Thomson mixture were replaced by *p*-nitrophenol. The absorption spectra of these solutions are compared in figure 12 to a commonly used glass filter (Chance ON–10). The NBS composite was prepared in dilute sulfuric acid and had a pH of 1. No fluorescence was observed under these conditions.

Over the range 250–600 nm, the NBS composite exhibits several broad maxima and minima whose absorbances are less dependent on spectral bandpass than the Chance ON–10 glass. Examples are given in figure 13 at two wavelengths for each.

The possible use of either Thomson solution or the NBS composite as an absorbance standard is limited by two factors, both of which arise from the presence of chromium (III) in these mixtures. First,

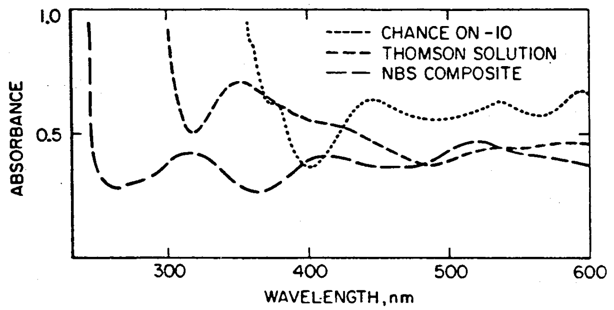


FIGURE 12. Absorption spectra of Thomas solution, an NBS composite and a Chance ON–10 glass.

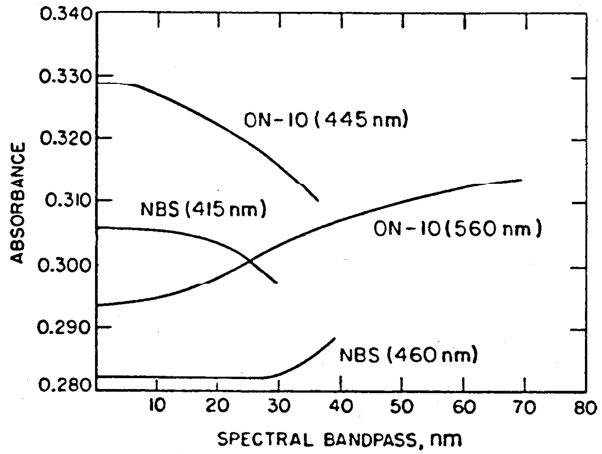
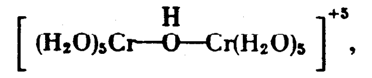


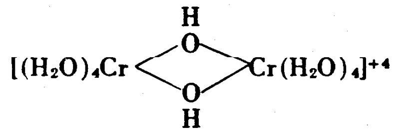
FIGURE 13. Variation of absorbance with spectral bandpass for NBS composite and Chance ON–10 glass.

an initial "aging" of 6–8 weeks period is needed in order to achieve adequate spectral stability at room temperature. Secondly, and even more critical for our purpose, is that the absorbances of the aged solutions exhibit a nonreversible temperature dependence. They must therefore always be stored at the temperature at which they were aged.

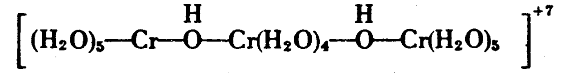
Hall and Eyring [35] have studied the constitution of chromic salts in aqueous solution and have suggested that during the aging process $Cr(H_2O)_6^{+3}$ ions polymerize to yield species of the type



$$\left[(H_2O)_5Cr{-}\overset{H}{O}{-}Cr(H_2O)_5\right]^{+5},$$



and



$$\left[(H_2O)_5{-}Cr{-}\overset{H}{O}{-}Cr(H_2O)_4{-}\overset{H}{O}{-}Cr(H_2O)_5\right]^{+7}$$

in which the observed increase in acidity is explained by the formation of —OH bridges. Recent studies [36] have shown that temperature, initial pH and the type of anion present significantly affect the equilibrium composition. Although refluxing can reduce the aging period from several months to several hours, this is of no practical advantage since the high temperature products, on cooling, are no longer stable.

In addition to the investigation of empirical mixtures, several water-soluble organic dyes have also been studied. Some typical absorption spectra are shown in figure 14. All of these dyes have high absorptivities and the concentrations used were of the order of 50 milligrams per liter.

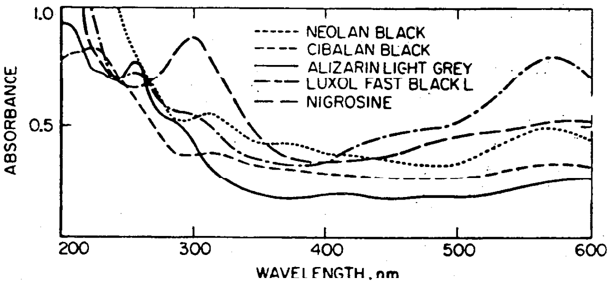


FIGURE 14. Absorption spectra of aqueous solutions of selected dyes.

Cibalan Black [37], Neolan Black [37] and Alizarin Light Grey [38] were selected for further evaluation. Aqueous solutions of these dyes were found insensitive to pH over the range 2–9, not affected by heat and relatively light fast. Solutions continuously exposed to fluorescent light exhibited an absorbance change of about 1 percent over a period of two months. These initial results appeared sufficiently promising to warrant purification and more extensive testing. Members of the Organic Chemistry Section have assisted in this effort. The purification of these dyes has proven extremely difficult, however, and yields of only a few hundred milligrams of each have been obtained. Much larger quantities are needed if these dyes are to be used as Standard Reference Materials. More efficient purification procedures are presently being sought.

D. Standard Reference Material (SRM) 931

The first liquid absorbance standard to be issued by the National Bureau of Standards is SRM 931. The preparation and certification of this Standard Reference Material are described below.

The filters were prepared by dissolving high purity cobalt and nickel in a mixture of nitric and perchloric acids. The weights of cobalt and nickel were chosen so that the absorbances of their aquo complexes were approximately equal at their absorption maxima. The absorbance of nitrate was adjusted to a comparable level by evaporation and subsequent addition

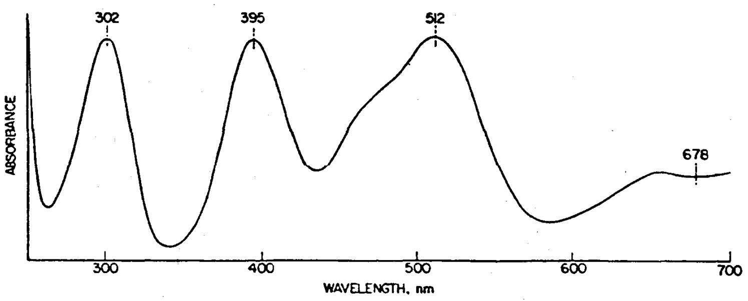


FIGURE 15. Typical absorption spectrum of SRM 931.

of small amounts of nitric acid. The final pH of these mixtures was about 1.

An absorption spectrum of a typical filter is shown in figure 15. The peaks at 302 and 512 nm are due to absorption by NO_3^- and $Co(H_2O)_6^{+2}$, respectively. The peak at 395 nm and the plateau at 650–700 nm are due to $Ni(H_2O)_6^{+2}$.

The filters are supplied in 10 ml ampoules which have been prescored for easy opening. An SRM unit consists of three sets of filters, each set containing a "Blank" (0.1N perchloric acid) and three absorbance levels of the filter. Each set is packaged in an individual tray for added convenience.

The filters were calibrated by randomly selecting ampoules from each lot. The certified absorbances are as follows:

TABLE 13. Certification of NBS SRM 931

Net absorbance[a]

Filter	Wavelength and (Bandpass), nm			
	302(1.0)	395(1.7)	512(2.0)	678(6.5)
"A" – 'Blank'	0.307 ± 0.003	0.304 ± 0.003	0.303 ± 0.003	0.115 ± 0.002
"B" – 'Blank'	0.608 ± 0.005	0.605 ± 0.005	0.606 ± 0.005	0.229 ± 0.003
"C" – 'Blank'	0.906 ± 0.007	0.907 ± 0.007	0.911 ± 0.007	0.345 ± 0.003

[a] Net absorbances "A" – 'Blank,' "B" – 'Blank,' and "C" – 'Blank' were determined using 10.00 mm cuvets at 25.0 °C.

The uncertainties are given at the 95 percent confidence level and include a possible systematic error of ±0.5 percent.

Absorbances are certified at 25.0 °C. Absorbances at other temperatures in the range 17 to 37 °C may be calculated using the formula:

$$A_t = A_{25}[1 + C_A(t-25)]$$

where: A_t = Absorbance at temperature t (°C)
A_{25} = Absorbance certified at 25.0 °C
C_A = Fractional change in absorbance per °C

The values of C_A at the four wavelengths are given below.

λ, nm	C_A
302	−0.0014
395	+0.0014
512	+0.0018
678	+0.0014

The absorbances of these filters will also depend on the spectral bandpass used. To insure that the measured absorbances do not differ by more than ±0.1 percent from the true values, the effective spectral bandpass should not exceed 1.5, 2.0, 3.3, and 8.5 nm at 302, 395, 512, and 678 nm, respectively.

IV. Conclusion

The availability of well characterized materials from which the analyst prepares his own solutions is preferable to the issuance of ampouled liquids. Although the former are not as convenient to use, they offer potentially greater accuracy because of their better stability.

The potassium dichromate and potassium hydrogen phthalate used in this study are issued by NBS as primary oxidimetric and acidimetric standards (SRM 136c and 84g, respectively). The potassium nitrate is presently issued as a thermal analysis standard (SRM 756). A product of comparable purity may be obtained by repeated recrystallization of reagent-grade material from distilled water. The cobalt and nickel solutions are best prepared from the high purity metals which are commercially available.

To certify absorptivities for these materials, one must resolve the question as to how well these systems obey Beer's law. For acidic potassium dichromate, the increased dimerization of $HCrO_4^-$ to $Cr_2O_7^{=}$ with increasing chromium concentration primarily accounts for the relatively large deviations observed. With potassium nitrate, one can only speculate that ionic interactions are enhanced at the higher concentrations. In neither instance are these deviations sufficiently large to limit the usefulness of these materials as liquid absorbance standards. Such deviations do require that the absorptivity data be used with proper precautions.

Additional studies are needed to explain the 0.2–0.3 percent deviations from Beer's law obtained for the various concentrations of alkaline chromate, potassium hydrogen phthalate, cobalt perchlorate and sulfate, and nickel perchlorate and sulfate. Although internal multiple reflection effects appear to be of the proper magnitude to account for these differences, initial attempts to measure such effects with magnesium fluoride-coated cuvets have not been successful. While extremely durable, magnesium fluoride reduces reflections by about 50 percent and more efficient coatings will probably be needed to resolve this problem. If internal reflections are responsible for the observed deviations, the absorptivity data given should not be corrected when using liquid absorbance standards for checking the accuracy of the photometric scale since these reflections are included in every measurement. However, in the determination of molar absorptivity, which is considered a fundamental property of the material, appropriate corrections should be applied. Until this question can be resolved, it is recommended that these materials be used at concentrations which yield an absorbance of at least 0.5 when measured in a 10 mm cuvet. With this restriction, it is believed that the uncertainty of the absorptivity data does not exceed ±0.5 percent at the 95 percent confidence level.

The authors wish to express their appreciation to Radu Mavrodineanu for providing the calibrated Schott filters and to David S. Bright for performing the computer calculations on the $HCrO_4^-/Cr_2O_7^{=}$ equilibrium.

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Considerations for the Use of Semi-Transparent Metallic Thin Films as Potential Transmittance Standards in Spectrophotometry*

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(May 20, 1976)

Various characteristics of evaporated metal-on-fused silica filters are discussed in relation to their optical transmission properties. Special metal holders provided with shutters were designed to be used with these filters, and are described in detail. Transmittance measurements, performed in various conditions, are reported and indicate that the evaporated metal-on-fused silica filters might present an acceptable material as transfer standards in spectrophotometry.

Key words: Evaporated metal-on-quartz; filters, transmittance; neutral filters; standard reference materials; transmittance characteristics; ultraviolet-visible filters.

The use of solid materials, in the form of glass filters and of solutions of inorganic and sometime organic compounds, to test the correct functioning of spectrophotometers is a common practice [1].[1] A variety of materials are available from the National Bureau of Standards (NBS) which can be used as transfer standards to verify the accuracy of the transmittance scale and the short and long term stabilities of conventional spectrophotometers. Two Standard Reference Materials (SRM's) have been issued by NBS for the verification of the accuracy of the transmittance scale; these are SRM 930 and SRM 931.

SRM 930 is a solid material which consists of a set of three glass filters having nominal transmittance of 10; 20; and 30 percent. They are certified for transmittance in the visible spectral range from 400 nm to 635 nm. A detailed description of this SRM is given in NBS Special Publication 260–51 [2]. SRM 931 is a liquid standard which consists of a solution of cobalt and nickel in dilute perchloric acid contained in glass ampoules [3]. The transmittance of these solutions is certified from 302 nm to 678 nm and should be used in conjunction with curvettes having a known light path; such curvettes are available from NBS as SRM 932 [4].

Both SRM's 930 (the glass filters) and 931 (the liquids) are limited in their spectral transmittance range and require the use of spectral bandpasses, from 2.2 nm to 6.5 nm for the glass filters and from 1.0 nm to 6.5 nm for the liquid filters depending of wavelength, when accurate transmittance values are sought.

The need to provide similar transfer standards, but with expanded spectral range to the ultraviolet and with less stringent spectral bandpass requirements, has resulted in an investigation to find adequate materials for this purpose. Such materials should fulfill the following conditions: (a) be transparent in the spectral range of interest, usually between 200 nm and 800 nm; (b) have a transmittance independent of wavelength (optically neutral); (c) have a spectral transmittance independent of temperature; (d) have low reflectance and be free of interferences; (e) be nonfluorescent; (f) be stable, homogeneous, and free of strain; (g) have mechanical stability for the size used (thickness, length, width) and be easy to fabricate by conventional techniques used in optical shops; (h) be simple to use in conjunction with the conventional spectrophotometers available today in analytical laboratories; (i) be readily available and relatively inexpensive. Thus, various solid materials were examined and the final choice was the evaporated metal-on-(non-fluorescent) fused silica type filter.

The transmission characteristics of such filters are illustrated in figure 1 and are compared with that of three glass filters and Ronchi ruling on glass.

The major limitation of the evaporated metal-on-quartz filters results from their intrinsic property of attenuating the incident radiation by reflecting rather than absorbing part of it. As a consequence of this property, this type of filter could generate stray radiations in the sample compartment of conventional spectrophotometers and is susceptible to produce interreflections when used with instruments equipped with lenses.

To determine the practical value of such filters as a transfer transmittance standard, comparative meas-

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[1] Figures in brackets indicate the literature references at the end of this paper.

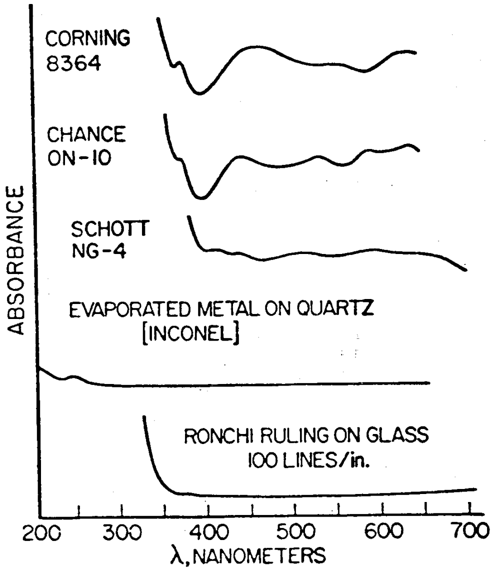

FIGURE 1. *Spectral characteristics for five transparent materials from 200 nm to 700 nm.*

urements were performed with the National Physical Laboratory in England (NPL), and the results are given in table I. In this case the inconel-on-fused silica filter was protected by a clean fused silica plate held in place with an organic cement. Except for a filter which showed some mechanical flaws in its structure, the reproducibility of transmittance measurements was as good as that obtained for the absorbing glass filters (table II). The error which would result from positioning the inconel-on-fused silica was measured by rotating from 0° angle to 3°—6°—and 9°. From the results obtained (table III) it can be concluded that in the case of the high-accuracy spectrophotometer used at NBS a positioning error within 3° can be tolerated.

The interreflection error affecting the measurements when a filter is inserted in the radiation path between the two lenses of a spectrophotometer was examined in detail [5]. The measured error for a glass filter established for the high-accuracy spectrophotometer was one or two 10^{-4} transmittance units; a value which is about four times larger was found for an evaporated metal-on-fused silica filter. It should be mentioned here that both the positioning and interreflection errors are, for a given material, instrument dependent.

As a result of these considerations a decision was made to produce a limited number of sets of evaporated inconel-on-fused silica and to test these filters in actual measurements performed on the conventional spectrophotometers manufactured in the United States.

One of the filters from a set is shown in figure 2. From left to right one can see the main body of the filter holder made from an aluminum alloy anodized black. This body is provided with dove-tail grooves which can accept a front and rear shutter made

TABLE I. *Comparison between the percent transmittances (%T) measured on three inconel-on-silica filters at NPL and NBS*

Wavelength nm	NBS, %T		NBS, %T average	NPL, %T
	1	2		
450.0	24.87	24.88	24.87$_5$	24.93
550.0	23.78	23.82	23.80	23.86
650.0	23.38	23.39	23.38$_5$	23.46
450.0	49.35	49.33	49.34	ᵃ 49.56
550.0	47.60	47.60	47.60	47.81
650.0	46.85	46.85	46.85	47.14
450.0	72.17	72.20	72.18$_5$	72.30
550.0	72.05	72.11	72.08	72.20
650.0	72.20	72.34	72.27	72.33

Average difference between NBS and NPL percent *T* values = −0.30 percent

ᵃ This filter had a flaw in the form of a crack which was sometimes visible and other times invisible. The larger differences found in the measurements of this filter may be due to this flaw.

TABLE II. *Comparison between the percent transmittances (%T) measured on three Schott NG-4 glass filters at NPL and NBS*

Wavelength nm	NBS, %T March 12, 1971	NBS, %T May 18, 1971	NBS, %T average	NPL, %T February 1971
440.0	12.92	12.91	12.91$_5$	12.93
465.0	14.96$_5$	14.98	14.97$_3$	15.01
590.0	11.70	11.64	11.67	11.67
635.0	12.72	12.68	12.70	12.72
440.0	19.62$_5$	19.58	19.60$_3$	19.62
465.0	22.38$_5$	22.35	22.36$_7$	22.43
590.0	19.06	18.95	19.00$_5$	19.01
635.0	20.45$_5$	20.37	20.41$_3$	20.47
440.0	32.89	32.86	32.87$_5$	32.98
465.0	35.52	35.54	35.53	35.66
590.0	31.16$_5$	31.10	31.13$_3$	31.21
635.0	32.56$_5$	32.52	32.54$_3$	32.62

Average difference between NBS and NPL percent *T* values = −0.19 percent

TABLE III. *Percent transmittance (%T) measured on a Schott neutral glass filter 2 mm thick, and an inconel-on-fused silica filter 2 mm thick at 590 nm, for normal incidence and for the angle of 3°; 6°; and 9°*

%T

Filter / Angle	Schott NG-4 Glass	Inconel-on-fused silica	
		Front	Back
0°	28.13	29.91	29.87
3°	28.10	29.98	29.87
6°	28.03	29.84	29.82
9°	27.98	29.92	29.90

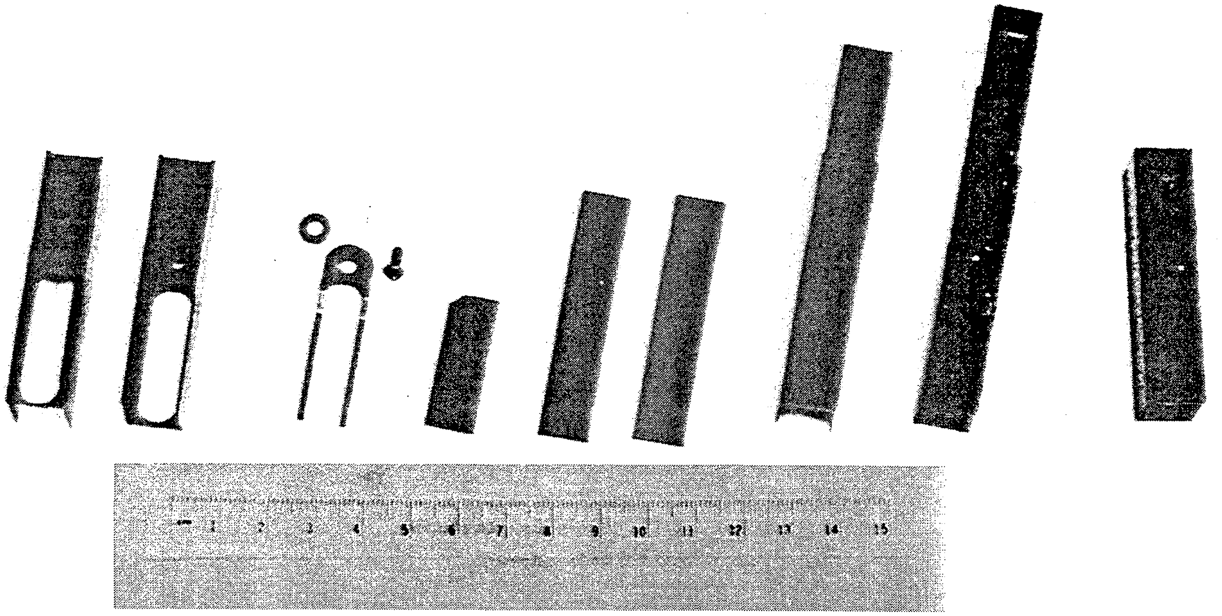

FIGURE 2. *Filter holder with shutters.*

From left to right: front and rear view of the holder body provided with dove-tail grooves; retaining spring with nylon screw and washer; filter; two shutters; front view of the filter holder with front shutter; rear view of the filter with rear shutter; filter holder with both shutters closed.

from a black plastic (Delrin [2]). The last three units illustrate the holder with these shutters on. A more detailed dimensional drawing of the filter holder is shown in figure 3. The shutters provide two functions: one is to protect the filter from contamination, the other to detect if stray radiant energy (SRE) is produced in the spectrophotometer compartment as a result of reflections generated by the incident beam on the semi-transparent metal layer. The determination of stray radiant energy generated in the sample compartment of the spectrophotometer, due to the reflecting propreties of these filters, may be assessed as follows: A background signal may be measurable when the instrument shutter at the photomultiplier tube is closed. Another slightly higher background signal may be detectable with the instrument shutter open and with the filter placed in the beam, in the sample compartment, with both sliding shutters closed. Both of these signals should be very small. A third signal may be detected when the front sliding shutter at the filter holder is removed and the rear sliding shutter is closed. If under these circumstances a signal is detected, it is very likely due to SRE produced by reflections resulting from the semi-transparent mirror which scatter from the walls of the cell compartment. This should also be low in magnitude. The transmittance of the filter is measured where both sliding shutters at the filter holder are removed.

Under these circumstances a source of systematic error can be from multiple reflections between the lenses in the instrument and the filter surfaces [5]. An indication of the existence and magnitude of such interreflection phenomena can be obtained by comparing the differences between the transmittance measurements and the certified values for SRM 930 and 931 to the corresponding differences for the inconel-on-fused silica filters.

Each set is made from three filters and one blank placed in individual metal holders which are provided with the front and rear sliding shutters. The selection of inconel as the semi-transparent metal layer was made on the basis of its relatively good optical neutrality in the spectral range from 250 nm to 700 nm. The fused silica substrate is of optical quality and non-fluorescent. All the silica filters, including the blank, were ground and polished at the same time and together to a parallelism of 0.02 mm and a flatness of less than 2 fringes (mercury 546.1 nm). The nominal dimensions are 30.5 mm by 10.4 mm by 2 mm thick, and the nominal transmittances of these filters are 1 percent; 20 percent; 30 percent; and 90 percent.[3] The transmittances of each set were measured at 250 nm, 300 nm, 340 nm, 400 nm, 440 nm, 465 nm, 500 nm, 546.1 nm, 590 nm, and 635 nm. Sets of these filters were sent to the manufacturers of spectrophotometers for evaluation and tests on the individual instruments. The results of these

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[2] In order to describe materials and experimental procedures adequately, it was occasionally necessary to identify commercial products by manufacturer's name or label. In no instance does such identification imply endorsement by the National Bureau of Standards, nor does it imply that the particular product or equipment is necessarily the best available for that purpose.

[3] The filter holders and the evaporated inconel-on-fused silica filters were made in the *NBS Instrument Shops.*

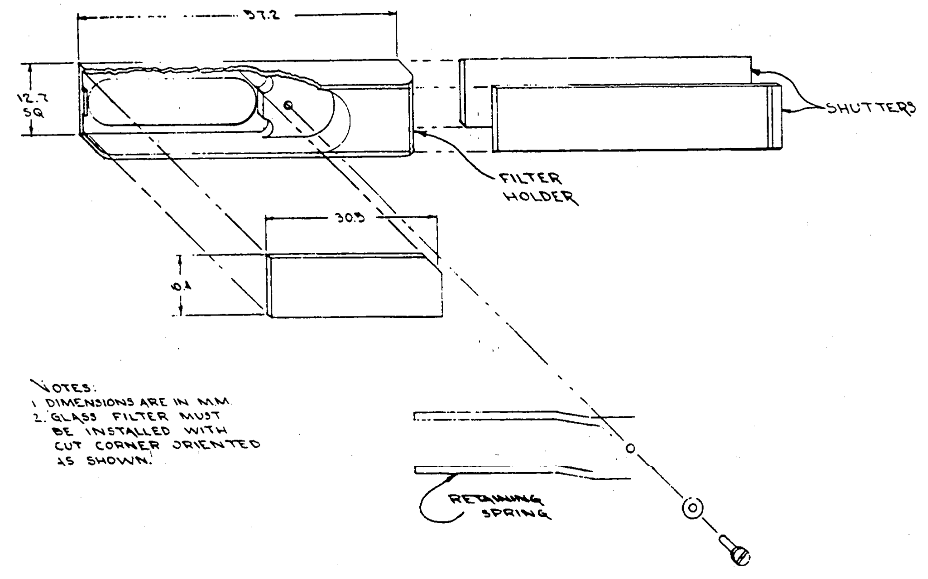

FIGURE 3. *Details of the filter holder with shutters.*

Nominal dimensions are in millimeters.

TABLE IV. *Percent transmittance (%T) measured on four inconel-on-fused silica filters at 10 wavelengths and on the NBS-IMR high-accuracy spectrophotometer and on two conventional instruments A and B*

	%T									
Wavelength nm / Instrument	250	300	340	400	440	465	500	546	590	635
NBS-IMR High-accuracy Spectrophotometer	1. 45	1. 81	1. 86	1. 94	2. 04	2. 13	2. 27	2. 49	2. 70	2. 90
	21. 39	21. 27	20. 33	18. 93	18. 33	18. 09	17. 90	17. 83	17. 86	17. 95
	28. 68	29. 92	30. 20	29. 96	29. 70	29. 55	29. 44	29. 44	29. 49	29. 57
	91. 42	92. 34	92. 66	92. 90	93. 01	93. 05	93. 11	93. 16	93. 19	93. 23
Spectrophotometer A	1. 53	1. 84	1. 93	2. 00	2. 12	2. 20	2. 34	2. 55	2. 75	2. 95
	21. 37	21. 24	20. 31	18. 91	18. 29	18. 07	17. 89	17. 82	17. 84	17. 92
	28. 67	29. 87	30. 19	29. 95	29. 70	29. 58	29. 49	29. 46	29. 52	29. 60
	91. 59	92. 60	93. 32	93. 20	93. 30	93. 30	93. 42	93. 46	93. 50	93. 58
Spectrophotometer B	1. 49	1. 85	1. 90	1. 97	2. 07	2. 16	2. 31	2. 53	2. 83	3. 06
	21. 6	21. 5	20. 5	19. 1	18. 5	18. 3	18. 1	18. 0	18. 3	18. 5
	28. 8	30. 0	30. 2	30. 0	29. 9	29. 7	29. 5	29. 5	29. 9	30. 0
	91. 4	92. 6	92. 6	93. 1	93. 3	93. 3	93. 3	93. 3	93. 9	93. 9

tests will permit to establish the usefulness of these filters as SRM's in spectrophotometry.

In the mean time comparative measurements were performed on two conventional spectrophotometers available at NBS, and the results are assembled in table IV. These results are preliminary measurements and should be considered with caution. The data seem to indicate, however, that the evaporated metal-on-fused silica filters may be an acceptable material for use as transfer standards in spectrophotometry.

NOTE.—The semi-transparent metallic surface of the filters described in this paper is not protected from environmental exposure and careless handling. A solution to this problem was found recently. It consists in placing on top of the fused silica plate carrying the semi-transparent evaporated metal layer a clear fused silica plate, about 1.5 mm thick, adequately ground and polished, to achieve an *optical contact* with the evaporated semi-transparent metal layer surface. The two fused silica plates are slightly beveled, and an optical cement is applied to protect the edges. Under these conditions the two fused silica plate assembly, with the semi-transparent evaporated metal layer in between, behaves optically as if it were a single semi-transparent plate, and provides full protection of the metal layer. Several sets of these new filters were made for evaluation in actual working conditions.

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Acidic Potassium Dichromate Solutions as Ultraviolet Absorbance Standards*

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(May 18, 1976)

The absorbances of five concentrations of potassium dichromate in 0.001 *M* perchloric acid have been determined at eight wavelengths in the ultraviolet on the National Bureau of Standards Institute for Materials Research high-accuracy spectrophotometer. Four of the wavelengths—235, 257, 313, and 350 nm—correspond to absorbance maxima or minima in the $HCrO_4^-$ spectrum and are useful wavelengths for checking the accuracy of the absorbance scale of narrow bandpass spectrophotometers. Although partial dimerization of $HCrO_4^-$ to $Cr_2O_7^=$ produces small positive deviations from Beer's law at these wavelengths, the apparent absorptivities calculated for each concentration are reproducible to one part in a thousand. The estimated uncertainties in the absorptivity values are ±0.7 percent at 0.1 absorbance (A) and ±0.2 percent near $A = 1$. These uncertainties include all known sources of possible systematic error and the 95 percent confidence level for the mean. The remaining four wavelengths used for measurement are near two predicted isosbestic points in the $HCrO_4^-/Cr_2O_7^=$ spectra. The absorptivities at 345 nm are sufficiently independent of concentration that this wavelength can be used for checking absorbance linearity to one part in a thousand over the range $A = 0.2–1$.

Key words: Absorbance linearity; accuracy; acidic potassium dichromate solutions; calibration of ultraviolet spectrophotometers; liquid filters; transfer standards; ultraviolet absorbance standards.

I. Introduction

At present, there are no certified standards available from the National Bureau of Standards (NBS) for checking the accuracy of the absorbance scale of spectrophotometers throughout the ultraviolet. The number of analytical applications in this important region of the spectrum, however, continues to increase relatively sharply and the need for such standards is becoming increasingly acute.

The area with perhaps the most serious need for ultraviolet absorbance standards at the moment is clinical chemistry. Workers in this field, for example, frequently use molar absorptivity as an index of purity of their biological standards. Only recently, Burnett [1] [1] has discussed the importance of accurate molar absorptivity measurements in the clinical laboratory. He especially emphasizes the need for spectrophotometric accuracy in clinical enzymology. Not only must the molar absorptivity of the substrate or enzyme-catalyzed reaction product be known accurately but also the individual absorbance measurements on the test sample must be accurate. This dual requirement for accuracy in this instance arises because high-purity and well-characterized enzyme preparations are not yet routinely available for use as standards.

A program has been under way at NBS in the Institute for Materials Research (IMR) since 1969 for the development and issuance of visible and ultraviolet transfer standards for use as Standard Reference Materials (SRM's). Two such standards are currently available: (1) SRM 930, consisting of a set of three neutral Schott NG glass filters and (2) SRM 931, an empirical inorganic solution available at three concentrations in 10-ml ampoules. Neither of these SRM's meets the present needs for absorbance standards in the ultraviolet. The glass filters do not transmit below about 350 nm while the inorganic solution is certified only at one wavelength in the ultraviolet (302 nm) and with an uncertainty of ±1 percent. Transfer standards capable of being certified throughout the range of 200–350 nm and with a smaller uncertainty are being investigated, however. The most promising solid filter at the moment for this purpose is the metal-on-quartz type. Some recent experiences with this filter are discussed in the paper immediately following [2]. Of the chemical or liquid-type filters that have been proposed, the weakly acidic potassium dichromate system is considered the best. A critical evaluation of this system, as well as several other candidate materials, was presented in a previous paper [3].

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*This work was supported in part by the National Institute of General Medical Sciences.

[1] Figures in brackets indicate the literature references at the end of this paper.

In this paper, we present absorptivity values for five concentrations of potassium dichromate in 0.001 *M* perchloric acid. These concentrations effectively span the absorbance range of $A = 0.1$–1.5 when measured in 10 mm cuvettes. The absorptivities were calculated from transmittance measurements at eight wavelengths in the ultraviolet on the IMR high-accuracy spectrophotometer. Four of the wavelengths—235, 257, 313 and 350 nm—correspond to absorbance maxima and minima in the $HCrO_4^-$ spectrum and are useful wavelengths for checking the accuracy of the absorbance scale of narrow bandpass spectrophotometers. Although partial dimerization of $HCrO_4^-$ to $Cr_2O_7^{=}$ produces small positive deviations from Beer's law at these wavelengths, the apparent absorptivities calculated for each concentration are reproducible to one part in a thousand. The estimated uncertainties in the absorptivity values are ± 0.7 percent at 0.1 absorbance (A) and ± 0.2 percent near $A = 1$. These uncertainties include all known sources of possible systematic error and the 95 percent confidence level for the mean. The remaining four wavelengths used for measurement are near two predicted isosbestic points in the $HCrO_4^-/Cr_2O_7^{=}$ spectra. The absorptivities at 345 nm are sufficiently independent of concentration that this wavelength can be used for checking absorbance linearity to one part in a thousand over the range $A = 0.2$–1.

II. Experimental Procedure

The potassium dichromate ($K_2Cr_2O_7$) used in this study was a special lot of analytical reagent grade material obtained from the J. T. Baker Chemical Company.[2] Drying studies performed at 105 °C indicated that the surface moisture of this material was less than 0.01 percent. No measurement of possible occluded water was made. However, a recent paper [4] by Yoshimori and Sakaguchi has shown that $K_2Cr_2O_7$ typically contains 0.01 to 0.02 percent occluded water which can be removed only by crushing and drying at 350 °C.

Coulometric assay of the undried potassium dichromate gave a purity, expressed as an oxidant, of 99.972 ± 0.005 percent at the 95 percent confidence level. Emission spectroscopy indicated that the only significant elemental impurities present were sodium and rubidium. Their concentrations were estimated to be 0.02 and 0.03 percent, respectively.

The concentrations of the $K_2Cr_2O_7$ solutions used throughout this study are expressed on a weight/weight basis. Milligram samples of $K_2Cr_2O_7$ were weighed to the nearest microgram on a microbalance. After dissolution in distilled water shown to be free of reducing impurities (see Discussion), 1 ml of 1 *M* perchloric acid was added and the solutions were diluted approximately to volume in 1-liter volumetric flasks. Each flask was fitted with a double-ribbed Teflon stopper (Kontes Glass Company, Vineland, New Jersey) to prevent evaporation. The weight of each solution was determined on a single-pan top-loading balance having a sensitivity of 0.01 g. NBS-calibrated weights were used to establish the accuracy of the balances. The concentrations of the solutions were then calculated after correcting all weights to vacuum. A solvent blank was prepared by diluting 1 ml of 1 *M* perchloric acid to 1 liter with distilled water.

The Institute for Materials Research high-accuracy spectrophotometer was used for performing the transmittance measurements which, in turn, were converted to absorbance. The design and construction of this instrument have been described in detail by one of us (RM) in reference [5] and will not be repeated here. Similarly, the quartz cuvettes used are also of NBS design and construction and have been described previously [6]. These cuvettes are currently available through the NBS Office of Standard Reference Materials as SRM 932. Each cuvette is certified for path length and parallelism to ± 0.0005 mm.

Prior to use the cuvettes were cleaned by soaking in concentrated (18 *M*) sulfuric acid for several hours. In order to minimize the heat of mixing, they were then transferred consecutively to 12, 6 and 3 *M* sulfuric acid before rinsing with distilled water. After rinsing thoroughly, the cuvettes were air-dried under an inverted Petri dish that served as a dust-protective cover.

For sample measurements, five calibrated cuvettes were placed in separate holders in the rotating sample compartment [5] of the IMR high-accuracy spectrophotometer and a reference filter (inconel-on-quartz) was placed in a sixth position. The cuvettes were left in their respective holders for the duration of the experiment. All transmittance/absorbance measurements were made relative to air in a temperature-controlled room at 23.5 ± 0.3 °C. The transfer of solvent blank and sample solutions to and from these cuvettes was made by means of borosilicate, Pasteur-type, disposable pipettes. After being rinsed with the test solution, a final transfer for measurement could be made in 10–15 seconds, after which time the cuvette was immediately stoppered with a snugly-fitting Teflon stopper.

Six sets of solutions having nominal concentrations of 20, 40, 60, 80 and 100 mg $K_2Cr_2O_7$/kg were prepared. Each concentration within a set was measured a minimum of six times at the eight wavelengths of interest. The absorbance, $A = -\log (T_{\text{Sample}}/T_{\text{Blank}})$, was computed for each wavelength and concentration from the average of the six transmittance measurements (T). Absorptivities were then calculated after correcting the absorbances for systematic errors due to cuvette path length, beam geometry and internal multiple reflections. No correction was applied for the reflections discussed by Mielenz and Mavrodineanu [7] from

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[2] In order to describe materials and experimental procedures adequately, it is occasionally necessary to identify the sources of commercial products by the manufacturer's name. In no instance does such identification imply endorsement by the National Bureau of Standards, nor does it imply that the particular product is necessarily the best available for that purpose.

internal components such as the lenses and slit jaws because these are adequately compensated for by the blank. A detailed account of these corrections and calculations will be given in an NBS 260 Special Publication which is now in preparation.

III. Discussion and Results

The ultraviolet absorbance spectrum of a 40 mg kg^{-1} solution of potassium dichromate in 0.001 *M* perchloric acid is shown in figure 1. The four wavelengths selected for certification of absorptivity of this absorbing system, namely 235, 257, 313 and 350 nm, are also indicated. The maxima and minima are sufficiently broad that serious restrictions are not placed upon instrumental spectral bandwidth requirements. The half bandwidths of the 257 and 350 nm peaks, for example, are approximately 60 nm so that an instrumental spectral bandwidth of 3 nm or less is sufficient for obtaining at least 0.999 of the maximum peak intensities.

The first experiment performed on the IMR high-accuracy spectrophotometer was the determination of the rinse behavior and reproducibility of the transmittance/absorbance measurements of the solvent blank. In the initial studies, measurements were made at 235 nm only, since any problems associated with the transfer of solution were expected to be the greatest at the shortest wavelength used. The results obtained for the five cuvettes employed are shown in figure 2. All exhibit a similar rinse pattern, attaining a minimum and constant absorbance value after 5–6 rinses (the term rinse as used here and throughout the remainder of this paper refers to the exchange of one cuvette volume for a second one). Once the blank measurements were in control, similar absorbance measurements were then made on the first series of $K_2Cr_2O_7$ solutions. Five concentrations having nominal concentrations of 20, 40, 60, 80 and 100 mg $K_2Cr_2O_7$ kg were measured. Table I summarizes the results of these measurements together with the final solvent blank values for 235 nm. The cycle of measurements shown here was then extended to the other seven wave-

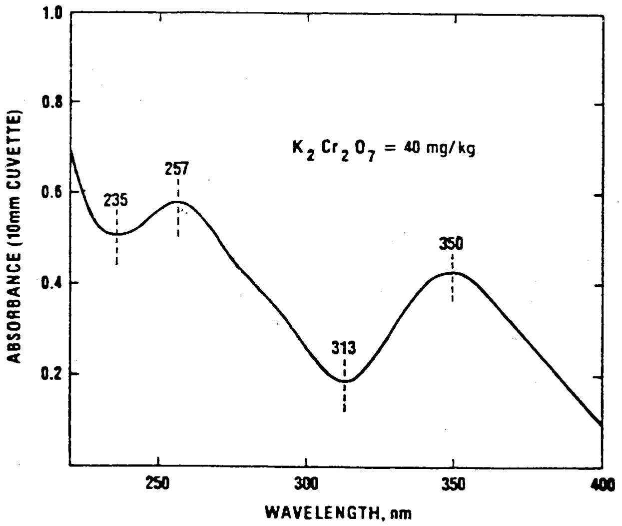

FIGURE 1. *Absorbance spectrum of* $K_2Cr_2O_7$ *in 0.001* M *perchloric acid.*

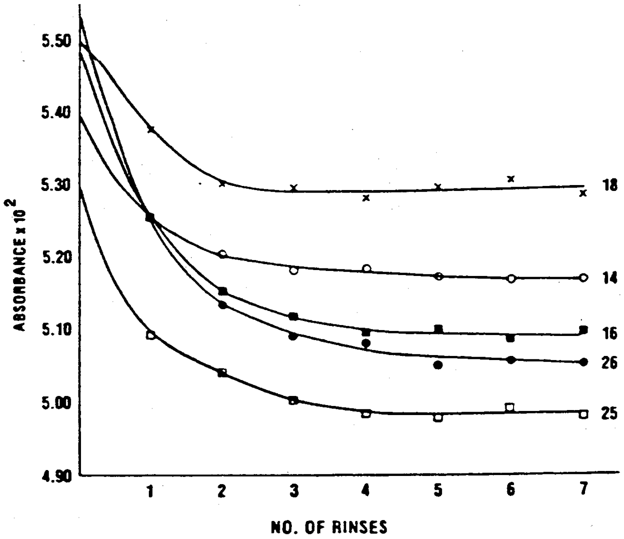

FIGURE 2. *Apparent absorbances of the blank solvent in the five curvettes used—Nos. 14, 16, 18, 25, and 26 ref.* [6].

TABLE I. *Reproducibility of absorbance measurements at 235 nm for a cycle of solvent and sample runs*

Cuvette No.	Solvent	Sample	Sample	Solvent
	After 5 rinses	After 5 rinses	After 7 rinses	After 5 rinses
14	0.05198	0.30128	0.30141	0.05213
16	.05099	.55052	.55057	.05093
18	.05310	.80066	.80074	.05321
25	.05002	1.04863	1.04855	.05022
26	.05064	1.30351	1.30333	.05065
Ref. filter	.55913	.55925	.55936	.55922

[a] Inconel-on-quartz.

lengths of interest. In all, six sets of $K_2Cr_2O_7$ solutions were measured. Typically, two volleys consisting of three transmittance/absorbance measurements were made on each of the 30 solutions. The first volley was made after rinsing the cuvettes, initially containing solvent, with five rinses of sample and was repeated again after two additional rinses. Since no systematic increase in absorbance was ever observed when the results of the first volley of measurements were compared to the second, the six measurements were invariably averaged. The absorbances were obtained by subtracting the solvent blank values from the sample readings. After applying appropriate corrections for beam geometry and internal multiple reflections, the corrected absorbances (A_{corr}) were used to calculate the desired absorptivities, using the relationship

$$\text{Absorptivity}=A_{\mathrm{corr}}/bc,$$

where b=internal light path in cm and c=concentration of $K_2Cr_2O_7$ solution in g kg^{-1}. The absorptivities computed for the five concentrations of $K_2Cr_2O_7$ solutions used are summarized in table II. The values tabulated were determined from least-squares plots of the experimental values and were subsequently normalized to the concentrations shown. The uncertainties given include all known sources of possible systematic error and the 95 percent (2σ) confidence interval for the mean. The random component of these uncertainties, based on standard deviations computed from residuals resulting from fitting the data to the various concentration levels for each wavelength, is 0.07 percent at the 2σ level.

It is observed that all absorptivities in table II increase with increasing $K_2Cr_2O_7$ concentration. These deviations from Beer's law are produced by the fact that, in weakly acidic media, chromium (VI) ions exist as two distinct absorbing species—$HCrO_4^-$ and its dimerization product, $Cr_2O_7^=$. The equilibrium between these two species may be represented as

$$2HCrO_4^- \overset{K_D}{\rightleftharpoons} Cr_2O_7^= + H_2O \qquad (1)$$

and the corresponding dimerization constant, K_D, is given by

$$K_D=\frac{[Cr_2O_7^=]}{[HCrO_4^-]^2} \qquad (2)$$

Although eq. (2) predicts that the formation of $Cr_2O_7^=$ is strictly a quadratic function of $K_2Cr_2O_7$ concentration, the value of $K_D=32.9$ (mol kg^{-1})$^{-1}$ obtained previously [3] is of such magnitude that the percentage of total chromium present as the $Cr_2O_7^=$ ion is very nearly a linear function of the $K_2Cr_2O_7$ concentration for the range of solutions studied. Calculated values of $HCrO_4^-/Cr_2O_7^=$ speciation in this system are given in table III.

TABLE III. $HCrO_4^-/Cr_2O_7^=$ *speciation in 0.001* M *perchloric acid solutions of* $K_2Cr_2O_7$.

$K_2Cr_2O_7$ Conc., g kg^{-1}	Percent Cr as $HCrO_4^-$	Percent Cr as $Cr_2O_7^=$
0. 020	99. 10	0. 90
. 040	98. 22	1. 78
. 060	97. 38	2. 62
. 080	96. 56	3. 44
. 100	95. 77	4. 23

The direction and magnitude of the expected deviations from Beer's law for the acidic potassium dichromate system can be ascertained when the speciation data in table III are combined with the spectral characteristics of the two chromium (VI) species. Figure 3 shows the relation of the absorbance spectra of the $HCrO_4^-$ and $Cr_2O_7^=$ ions. At wavelengths of 235, 257, 313 and 350 nm, the $Cr_2O_7^=$ ion is shown always to have a larger absorptivity than the $HCrO_4^-$ ion. Hence, the measured absorptivities at these wavelengths should increase with increasing $K_2Cr_2O_7$ concentration.

From the same argument, the data in figure 3 also suggest that there are two wavelengths near 320 and 345 nm where Beer's law is obeyed. In an attempt to determine these isosbestic points experimentally, absorbance measurements were also made on the

TABLE II. *Absorptivities of* $K_2C_2O_7$ *in 0.001* M *perchloric acid at 23.5 °C*

Absorptivity, kg g^{-1}cm^{-1}

$K_2Cr_2O_7$ Conc., g kg^{-1}	235(1.2)[b]nm	257(0.8)nm	313(0.8)nm	350(0.8)nm	Uncertainty [c]
[a]0. 020	12. 243	14. 248	4. 797	10. 661	0. 034
. 040	12. 291	14. 308	4. 804	10. 674	[d]. 022
. 060	12. 340	14. 869	4. 811	10. 687	[d]. 020
. 080	12. 388	14. 430	4. 818	10. 701	[d]. 020
. 100	12. 436	14. 491	4. 825	10. 714	[d]. 019

[a] Nominal concentration; all weights corrected to vacuum.

[b] Wavelength and, ( ), spectral bandwidth.

[c] Includes estimated systematic errors and the 95 percent confidence interval for the mean.

[d] For wavelength of 313 nm, the uncertainty is reduced to half of these values for $K_2Cr_2O_7$ concentrations of 0.040, 0.060, 0.080 and 0.100 g kg^{-1}.

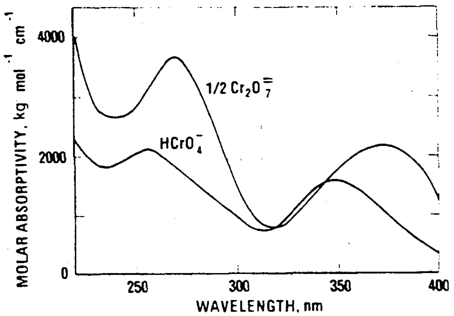

FIGURE 3. *Absorbance spectra of the* $HCrO_4^-$ *ion and its dimerization product,* $Cr_2O_7^=$

same solutions used above at 322, 323, 345 and 346 nm. The corresponding absorptivity values are given in table IV. Although small systematic deviations from Beer's law are still present, the absorptivities calculated for 345 nm are sufficiently constant that the acidic $K_2Cr_2O_7$ system can be used at this wavelength, over the concentration range shown, to check the absorbance linearity of narrow bandpass spectrophotometers to one part in a thousand.

The variation of absorptivity of the acid $K_2Cr_2O_7$ system with temperature is the smallest that we have observed for any liquid filter that we have studied to date. For the four wavelengths recommended for checking the accuracy of the absorbance scale—235, 257, 313, and 350 nm—the absorptivities decrease with increasing temperature. Over the range 20–30 °C, the corrections are, respectively, −0.05, −0.05, −0.02, and −0.05 percent per degree Celsius. The −0.02 percent correction found at 313 nm corresponds closely to the correction predicted from the thermal expansion of the solvent. Until further evaluation can be made of the temperature dependence of 345 nm wavelength recommended for checking absorbance linearity, measurements should be restricted to 23.5 ± 1 °C.

Several considerations were involved in the selection of perchloric acid for acidifying the $K_2Cr_2O_7$ solutions. First, perchloric acid was preferred over the sulfuric acid which has been used traditionally because perchlorate ion, unlike sulfate, has no tendency to form mixed complexes with chromium (VI) species. Secondly, the choice of 0.001 *M* acid rather than 0.01 *M* was based on two factors: (1) the 0.001 *M* acid represented a practical compromise for minimizing conversion of $HCrO_4^-$ to either H_2CrO_4 or $CrO_4^=$ and (2) the lower acid concentration substantially lowered the oxidation potential (~13 percent) of the chromium (VI) ions and hence improved the solution stability of the proposed $K_2Cr_2O_7$ standards.

A final point that needs to be emphasized concerns the quality of the distilled water used for preparing the standard solutions. Acidic potassium dichromate is a potential oxidant and consequently the distilled water used must be shown to be free of reducing impurities in order that the absorptivity data given in this paper be valid. A simple but yet definitive test is outlined in figure 4. In this study a 500-fold dilution of a 100 mg kg⁻¹ standard solution of $K_2Cr_2O_7$ was made with the distilled water in question and its absorbance measured at 350 nm. Not only did the measured absorbance agree with the predicted value, thus indicating no reduction of chromium (VI), but also subsequent measurements showed that this $K_2Cr_2O_7$ solution standard should be stable for at least two months provided it is adequately protected against evaporation. In instances where this test shows the distilled water to be suspect, it is recommended that the water be redistilled from alkaline potassium permanganate in order to preoxidize the impurities.

Finally, the general use of solid and liquid transfer standards for calibrating the absorbance scale of a precision commercial spectrophotometer is illustrated in figure 5. The measurements on the glass

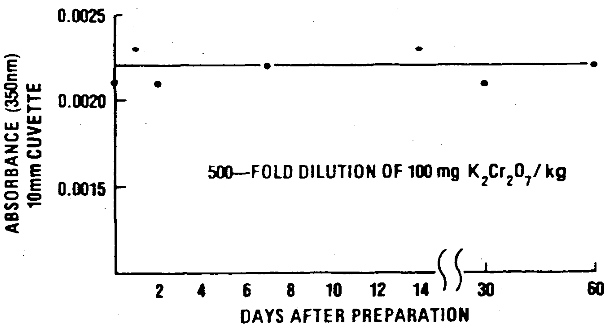

FIGURE 4. *Test of the distilled water for reducing impurities.*

TABLE IV. *Absorptivities of* $K_2Cr_2O_7$ *in 0.001* M *perchloric acid near two predicted isosbestic wavelengths; temperature 23.5 °C*

Absorptivity, kg g⁻¹ cm⁻¹

$K_2Cr_2O_7$ Conc., g kg⁻¹	322 (0.8)[b] nm	323 (0.8) nm	345 (0.8) nm	346 (0.8) nm
[a] 0. 020	5. 845	6. 065	10. 593	10. 615
. 040	5. 842	6. 062	10. 595	10. 621
. 060	5. 838	6. 059	10. 598	10. 627
. 080	5. 835	6. 056	10. 600	10. 632
. 100	5. 831	6. 053	10. 602	10. 638

[a] Nominal concentration; all weights corrected to vacuum.
[b] Wavelength and, ( ), spectral bandwidth.

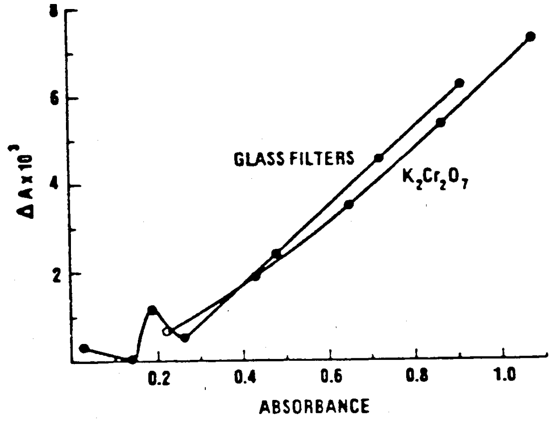

FIGURE 5. *Correction* ΔA *for the absorbance scale of a precision commercial spectrophotometer.*

filters were performed at 440 nm while the $K_2Cr_2O_7$ measurements were made at 350 nm. At both wavelengths the absorbances measured on the precision spectrophotometer are high and should be corrected by subtracting the appropriate ΔA values.

IV. Conclusion

Following widespread use in the collaborative testing of spectrophotometers for more than 25 years, the absorptivities of the acidic potassium dichromate system have now been determined with a known accuracy from measurements performed directly on the NBS Institute for Materials Research high-accuracy spectrophotometer. Later this year, crystalline $K_2Cr_2O_7$, together with detailed instructions on preparing solutions from it, will be available from NBS through its Office of Standard Reference Materials as an SRM. In conjunction with the calibrated quartz cuvettes previously issued (SRM 932), it is believed that this material will provide a valid check of the accuracy of the absorbance scale of narrow bandpass spectrophotometers in the ultraviolet from 235–350 nm.

The authors gratefully acknowledge the assistance of the following persons at NBS during various phases of this work: K. D. Mielenz, for helpful discussions of fundamental spectrophotometric parameters; G. Marinenko, for performing the coulometric assay; J. A. Norris, for the the emission spectrographic analyses; H. H. Ku, for the statistical analysis of the data; and R. W. Seward, for supplying through the Office of Standard Reference Materials the samples of potassium dichromate.

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(Paper 80A4–908)

U.S. Department of Commerce
Juanita M. Kreps
Secretary
National Bureau of Standards
Ernest Ambler, Acting Director

National Bureau of Standards Certificate

Standard Reference Material 930D

Glass Filters for Spectrophotometry

R. Mavrodineanu and J. R. Baldwin

This Standard Reference Material is intended as a reference source for the verification of the transmittance and absorbance scales of spectrophotometers. It consists of three individual glass filters in their metal holders and one empty filter holder. The holders are provided with shutters to protect the glass filters when not in use. These shutters must be removed at the time of measurements and be replaced after the measurements have been completed. Each filter bears an identification number, and the upper left corner has been removed to indicate correct placing in the metal holder.

Filter & Set Ident. Number	TRANSMITTANCE (T)					TRANSMISSION DENSITY ($-\log_{10}T$)				
	Wavelength and (Bandpass) nm					Wavelength and (Bandpass) nm				
	440.0 (2.2)	465.0 (2.7)	546.1 (6.5)	590.0 (5.4)	635.0 (6.0)	440.00 (2.2)	465.0 (2.7)	546.1 (6.5)	590.0 (5.4)	635.0 (6.0)

Date of Certification:

The transmittance values (T) can be converted to percent transmittance (%T) by multiplying by 100. The transmission densities are calculated from the measured transmittance (T). These values should be indicated by the absorbance scale of the spectrophotometer if the filters are measured against air. The transmittance values given were measured against air at an ambient temperature of 23.5 °C.

The transmittance values are estimated to be accurate to within ±0.5 percent at the time of certification. This uncertainty includes the effects of the random and systematic errors of the calibration procedure, as well as possible transmittance changes of the filters during the period of calibration.

Aging of the glass may cause some filters to change transmittance by about ±1 percent over a period of approximately one year from the date of calibration. Improper storage or handling of the filters may also cause changes [5]. It is recommended that the filters in the holders be handled only by the edges with soft plastic (polyethylene) gloves and optical lens tissue. When not in use they should be stored in their holders and in the container provided for this purpose. Extended exposure to laboratory atmosphere and dusty surroundings should be avoided. In cases where verification is desirable, the filters should be returned to the National Bureau of Standards for cleaning and recalibration.

This Standard Reference Material was issued with the assistance and advice of K. D. Mielenz and J. R. DeVoe.

The technical and support aspects involved in the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by T. W. Mears and R. W. Seward.

Washington, D.C. 20234
August 1, 1977

J. Paul Cali, Chief
Office of Standard Reference Materials

The transmittance measurements were made using the high-accuracy spectrophotometer designed and built at the NBS Institute for Materials Research [1]. This instrument represents the primary transmittance standard; its transmittance accuracy was established using the double-aperture method of linearity testing [1,3,5,6]. The estimated standard deviation obtained from a set of 20 measurements of the high-accuracy spectrometer used for transmittance measurements on glass filters of the SRM 930 type is 5×10^{-5} transmittance units for transmittance between 0.1 and 0.3. The bounds of the systematic error of the instrument are estimated to be $\pm 10^{-4}$ transmittance units.

The neutral glasses for the filters were provided by Schott of Mainz, Germany, and are designated as "Jena Color and Filter Glass." The glass material was prepared by the manufacturer especially for SRM 930. It was fine-annealed and was selected for best homogeneity and minimum of inclusions and striae. The glass filters were aged at NBS for about one year prior to certification. They are of the type NG-4 and NG-5, and their nominal transmittances are 10, 20, and 30 percent [2,5]. The glasses were selected to provide a means to verify the transmittance scale of conventional spectrophotometers at three levels. The exposed surface of the glass is approximately 29 x 8 mm, measuring from a point 1.5 mm above the base of the filter holder (see figure). The empty filter holder is provided to be used in the reference beam of the spectrophotometer so that approximately equivalent conditions of stray radiations are achieved for both beams.

The transmittance of the filters depends upon the intrinsic properties of the material, wavelength, spectral bandpass, geometry of the optical beam, temperature, and positioning of the filter. A change of ambient temperature of 1 or 2 °C from 23.5 °C will not significantly affect the calibration [5]. Changes in the transmittance may be caused by surface conditions, aging of the glass, exposure to a harmful atmosphere, or careless handling as indicated on the face of this certificate [2,3,4,5]. The effective spectral bandpasses used to determine the certified values are given on the face of the certificate and the transmittance measurements are made by producting the vertical image of the slit (about 8 mm by 0.5 mm), using a convergent beam geometry with an aperture ratio f:10, in the middle of the entrance face of the glass filter. The filters were measured in the spectrophotometer in a position perpendicular to the incident light beam as shown in the attached figure.

SRM 930D is stored in a black anodized aluminum container provided with a threaded cap made of the same metal. Each filter is placed in a cylindrical cavity to prevent any contact between the filter face and the walls of the storage container. Contamination of the glass filter surface with particulate matter due to static charges is minimized through the metallic nature of the container. Each filter holder is provided with a flat leaf spring which is inserted into the cylindrical cavity to minimize damage during transportation. These springs can be removed during normal use in the laboratory.

Prior to certification measurements, each filter was examined for surface defects and thorougly cleaned [5]. Should the surface of the glass filter become contaminated, no attempt should be made to clean it unless the user has the facilities to demonstrate that the cleaning treatment has not altered the glass surface or degraded the accuracy of the certified values. As the Standard Reference Material is a transfer standard, the only means available to verify its integrity is remeasuring its transmittance with a primary standard instrument similar to that used in this certification [1,4,5]. In most cases where verification, or recertification, of the transmittance values is desirable, it will be most expeditious to return the filters to the National Bureau of Standards for measurement. Prior to shipment, the NBS Office of Standard Reference Materials should be contacted regarding the conditions and cost of verification and/or recertification.

We wish to acknowledge the cooperation of George N. Bowers, Jr., M.D., of Hartford Hospital, Hartford, Connecticut; Royden N. Rand, Ph.D., of the Eastman Kodak Co. Research Laboratories, Rochester, New York; and Donald S. Young, M.D., Ph.D., of the National Institutes of Health, Bethesda, Maryland.

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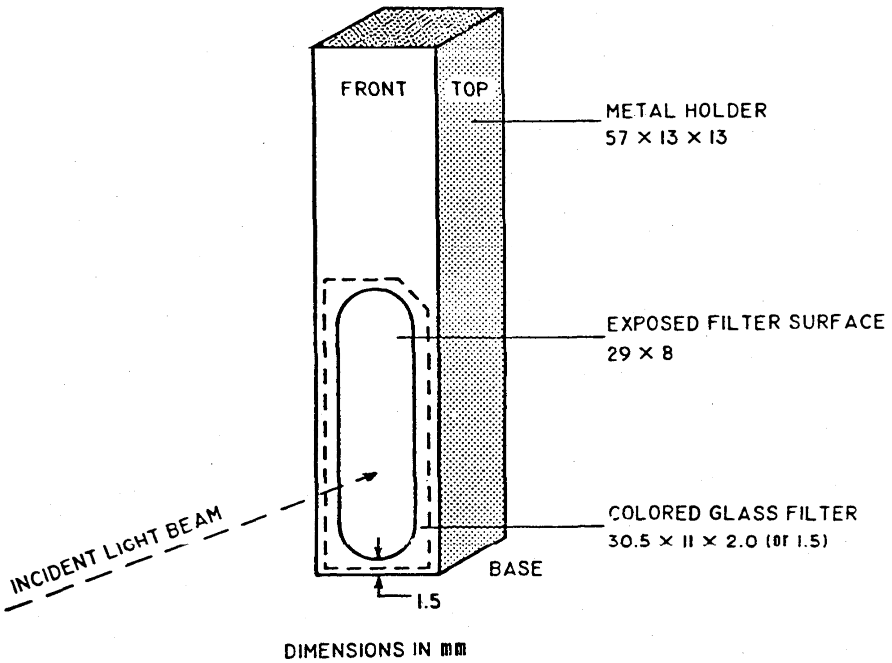
METAL HOLDER FOR THE COLORED GLASS FILTERS*



*Shutters not illustrated.

U. S. Department of Commerce
Malcolm Baldrige
Secretary

National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards
Certificate
Standard Reference Material 931c
Liquid Absorbance Standards for Ultraviolet and Visible Spectrophotometry

This Standard Reference Material is certified as solutions of known net absorbances at specific spectral wavelengths. It is intended primarily for use in the calibration and checking of accuracy of the photometric scale of narrow bandpass spectrophotometers and for routine critical evaluation of daily working standards used in spectrophotometry. This Standard Reference Material is applicable for calibrating those instruments that provide an effective spectral bandpass of 1.5 nm or less at 320 nm, 2.0 nm or less at 395 nm, 3.3 nm or less at 512 nm, and 8.5 nm or less at 678 nm [1].

Filter	Net Absorbance[a] Wavelength and (Bandpass), nm			
	302(0.8)	395 (0.8)	512(0.8)	678(0.8)
"I" -"Blank"	0.305 ± 0.001	0.309 ± 0.001	0.307 ± 0.001	0.116 ± 0.001
"II" -"Blank"	.608 ± .002	.612 ± .002	.609 ± .002	.232 ± .001
"III"-"Blank"	.938 ± .002	.912 ± .002	.904 ± .002	.346 ± .001

[a] Net absorbances ("I" - "Blank", "II" - "Blank", and "III" - "Blank") were determined using 10.00-mm cuvettes (SRM 932) at 25 °C. See Instructions for Use.

The transmittance measurements leading to the certification of this SRM were performed on the high accuracy spectrophotometer located in the NBS Center for Analytical Chemistry. The design and construction of this instrument have been described previously [2]. This instrument is the primary transmittance standard; its accuracy has been verified using the double aperature radiation-addition principle. The bounds of systematic error of this instrument are estimated to be $\pm 10^{-4}$ transmittance units.

These liquid absorbance filters were calibrated at the wavelengths and conditions indicated by measuring the transmittance, T, of the "Blank" and solutions "I", "II", and "III" against air as a reference. The values of T were used to calculate the corresponding apparent absorbances, A_a, using the relationship $A_a = -\log_{10} T$. The certified net absorbances were obtained by subtracting the apparent absorbances of the "Blank" solution from the apparent absorbances calculated for solutions "I", "II", and "III". The uncertainties of the certified values include all known sources of possible systematic error and the 95 percent confidence level for the mean.

While no long-term stability studies have been made on this lot (931c), studies on previous lots (931, 931a, and 931b) over three-year periods showed no degradation of the material when stored in the original sealed ampoules. Therefore, this material is certified only for use within three years following the date of purchase.

The preparation of the filters was performed by E.R. Deardorff, Inorganic Analytical Research Division, and the transmittance measurements were performed by M.V. Smith, Office of Standard Reference Materials. Technical leadership for the preparation and measurements leading to certification was provided by R.W. Burke, Inorganic Analytical Research Division.

The overall direction and coordination of technical measurements leading to certification were performed under the chairmanship of E.L. Garner.

The technical and support aspects involved in the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by R.W. Seward.

Washington, D.C. 20234
February 12, 1982

George A. Uriano, Chief
Office of Standard Reference Materials

Temperature Dependence

Absorbances at various temperatures (17 to 35 °C) may be calculated using the equation

$$A_t = A_{25}[1+C_A(t-25)]$$

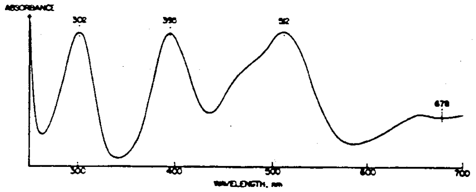
where: A_t = Absorbance at temperature t (°C)
A_{25} = Absorbance certified at 25.0 °C
C_A = Fractional change in absorbance per °C

The values of C_A, at the four wavelengths, are given below. [NOTE: At wavelength 302 nm, absorbance decreases with increasing temperature; at the other wavelengths, absorbance increases with increasing temperature.]

Wavelength, nm	C_A
302	−0.0014
395	+0.0014
512	+0.0018
678	+0.0014

Preparation of Filters

The filters were prepared by dissolving high-purity cobalt and nickel in a mixture of nitric and perchloric acids. The absorbance spectrum of the resulting solution is shown in the following figure. The maxima at 302 and 512 nm are due to absorbance by NO_3^- and $Co(H_2O)_6^{++}$ respectively. The maximum at 395 nm and the plateau at 650-700 nm are due to $Ni(H_2O)_6^{++}$ The pH of these solutions is about 1.



Instructions for Use

This material is for use as a spectrophotometric absorbance standard.

1. Select two clean 10.00 mm cuvettes free of scratches. At least one should be fitted with a ground glass or Teflon stopper to minimize evaporation. Reserve it for all sample measurements.
2. Mark each of the cuvettes to assure the same orientation in the spectrophotometer.
3. Place the cuvettes in their respective holders and fill with distilled water. (Borosilicate Pasteur-type pipettes fitted with rubber bulbs are recommended for transferring all solutions to and from the cuvettes. Soft glass pipettes, which are available commercially, contain residual amounts of ultraviolet absorbing material, but may be used after proper cleaning. Several rinses, first with isopropyl alcohol and then with distilled water, are generally adequate.
4. Obtain the optical mismatch of the cuvettes at 302, 395, 512, and 678 nm, using the spectral bandpass limitations given on the face of the certificate.
5. Empty the cuvettes by suction without removing them from their holders, refill with distilled water and measure the absorbances again at each of the above wavelengths.

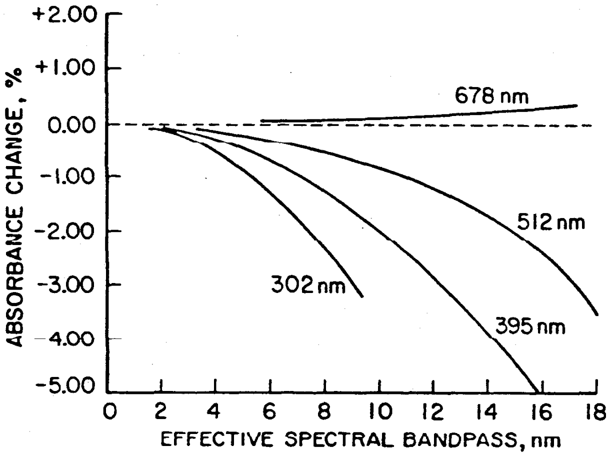
6. Repeat the emptying and refilling operation until constant absorbance readings are obtained.
7. Using the liquid filters provided, measure, in turn, the absorbance of the "Blank", "I", "II", and "III" against distilled water. Shake each ampoule before opening to remix any condensate which may have been collected in the neck. (The ampoules have been prescored directly below the gold band to facilitate opening.)
8. Subtract the appropriate "Blank" reading from the absorbances obtained for "I", "II", and "III". These net absorbances should agree with the certified values within the uncertainties specified. Consult the manufacturer of the instrument if they do not.

The above instructions are for use with the standard 10-mm rectangular cuvette. For calibration of the several spectrophotometric systems used in various automated instruments, the user is referred to the instruction manual for the particular instrument.

The absorbances of these liquid absorbance standards will depend not only on the accuracy of the photometric scale, but also on the wavelength accuracy and the spectral bandpass. A mercury lamp is recommended for checking the wavelength scale. In addition, for those spectrophotometers having a hydrogen (H) or deuterium (D) source, the two emission lines at 486.1 and 656.3 nm (H) or 486.0 and 656.1 nm (D) may provide a convenient check at these wavelengths.

To ensure that the measured absorbances are not significantly different from the certified values, the following restrictions are placed on the size of the spectral bandbass selected: To obtain ±0.1 percent of the true value, the effective spectral bandpass should not exceed 1.5, 2.0, 3.3, and 8.5 nm at 302, 395, 512, and 678 nm, respectively.

For ±0.2 percent, the respective bandpasses should not exceed 2.2, 2.9, 4.8, and 12.3 nm. Additional information on the effect of spectral bandpass on the absorbances of these filters is given in the figure below. These curves are not to be used, however, to correct the measured absorbances.



This Standard Reference Material should be kept in the original sealed ampoules. Once opened, the material should be used immediately. No attempt should be made to reseal the ampoule. In addition, it is recommended that this Standard Reference Material not be used after three years from the purchase date.

References

[1] R. W. Burke, E. R. Deardorff, and O. Menis, J. Research, Nat. Bur. Stand. 76A, 469-482 (1972).

[2] R. Mavrodineanu, J. Research, Nat. Bur. Stand. 76A, 405-425 (1972).

Note: The above papers are also published in NBS Special Publication 378, Accuracy in Spectrophotometry and Luminescence Measurements, R. Mavrodineanu, J. I. Shultz, and O. Menis, Editors, U.S. Government Printing Office, Washington, D.C. 20402, 1973.

U. S. Department of Commerce
Philip M. Klutznick
Secretary
National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards

Certificate

Standard Reference Material 932

Quartz Cuvette for Spectrophotometry

R. Mavrodineanu and J. W. Lazar

This Standard Reference Material consists of a single, accurately calibrated cuvette that is issued for use in the production of accurate spectrophotometric data on liquids. The design and nominal dimensions of the all-quartz cuvette are shown in Figure 1. The pathlength of the cuvette is defined by the distances between the two optically transparent windows measured at several heights within the cuvette. Cuvettes issued as Standard Reference Material 932 have a nominal pathlength of 10 mm. The pathlength and parallelism are certified, at the time of measurements, with an uncertainty of ±0.0005 mm as determined by measurements (at 20 °C) taken at the positions indicated below.

Cuvette number   is issued with this certificate. For this cuvette the following measurements were obtained:

Height mm	Pathlength mm

Date of Certification:

The overall direction and coordination of the technical measurements leading to certification were performed under the chairmanship of O. Menis and J. A. Simpson.

The technical and support aspects involved in the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by T. W. Mears.

Washington, D.C. 20234
December 2, 1980
(Revision of certificates
dated 11-5-73, 6-16-77, and 8-2-78)

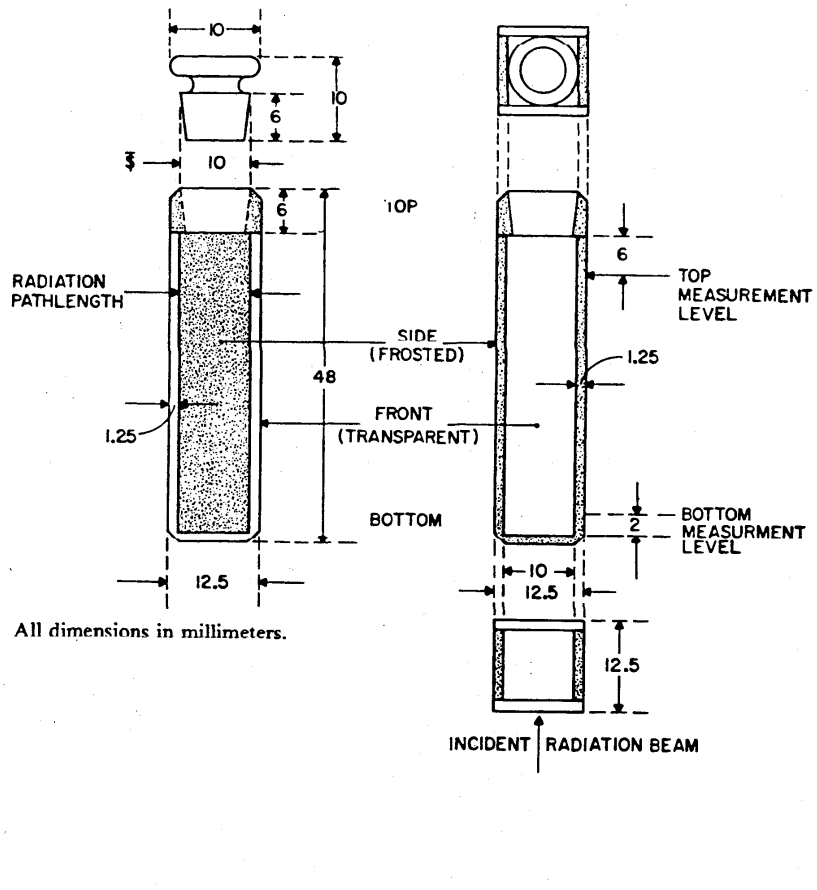
George A. Uriano, Chief
Office of Standard Reference Materials

The original cuvettes were designed and produced at the National Bureau of Standards (NBS) using special techniques and non-fluorescent optical-quality fused quartz as described in NBS SP 260-32[1]. The transparent windows are attached to the body of the cuvette by direct fusion, and the unit was stress-relived by annealing. The radiation pathlength measurements of the cuvette were performed using electronic feeler-gauge type instruments capable of a resolution of 5 parts in 10^6. To preserve the integrity of the certified values, the cuvette must be handled with great care and should be held only by the frosted-quartz side windows. When not in use, it should be stored in the container provided for this purpose. Extended exposure to laboratory atmosphere and dusty surroundings is to be avoided. Improper handling of the cuvette and the use of solutions that can corrode or errode the quartz could degrade the certified pathlength values. In cases where verification of the pathlength is desirable, the cuvette should be returned to NBS for examination and, if needed, recalibration. Prior to shipment the NBS Office of Standard Reference Materials should be contacted regarding the conditions and cost of the verification and/or recertification.

The development and production of SRM 932 is a result of the combined efforts of the Center for Analytical Chemistry, the Center for Mechanical Engineering and Process Technology, and the Instrument Shops Division. The radiation pathlength measurements of all of these cuvettes have been performed, at NBS, by E. G. Erber of the Mechanical Processes Division.

Reference:

[1] Mavrodineanu, R., and Lazar, J. W., Standard Reference Materials; Standard Quartz Cuvettes for High Accuracy Spectrophotometry, Nat. Bur. Stand. (U.S.), Spec. Publ. 260-32 (December 1973). Superintendent of Documents, U.S. Government Printing Office, Washington, D.C. 20402, 55 cents.



All dimensions in millimeters.

U.S. Department of Commerce
Juanita M. Kreps
Secretary

National Bureau of Standards
Ernest Ambler, Acting Director

National Bureau of Standards
Certificate

Standard Reference Material 935

Crystalline Potassium Dichromate
for Use as an Ultraviolet Absorbance Standard

R. W. Burke and R. Mavrodineanu

This Standard Reference Material consists of crystalline potassium dichromate of established purity. Solutions of known concentrations of this SRM in 0.001 N perchloric acid are certified for their apparent* specific absorbances**, ϵ_a, at 23.5 °C.

This SRM is intended to be used as a reference standard for the verification of the accuracy and linearity of the absorbance scale of absorption spectrometers that can provide an effective spectral bandpass of 1.2 nm or less at 235 nm, and 0.8 nm or less at 257, 313, 345, and 350 nm. Such verification is accomplished by comparing the measured apparent absorbances, A_a, to the A_a calculated from the certified ϵ_a values as described under "Instructions for Use."

Table 1 gives the certified values of ϵ_a in $kg \cdot g^{-1} \cdot cm^{-1}$ for five concentrations of the SRM 935 potassium dichromate in 0.001 N perchloric acid at 23.5 °C and the indicated wavelengths and spectral bandpasses for a 1-cm internal pathlength.

Table 1. ϵ_a, Apparent Specific Absorbance,[a] $kg \cdot g^{-1} \cdot cm^{-1}$

Nominal Concentration $g \cdot kg^{-1}$	Wavelength and (Bandpass) nm					
	235.0(1.2)	257.0(0.8)	313.0(0.8)	345.0(0.8)[b]	350.0(0.8)	Uncertainty[c]
0.020	12.260	14.262	4.805	10.604	10.672	± 0.034
.040	12.304	14.318	4.811	10.603	10.682	± .020[d]
.060	12.347	14.374	4.816	10.602	10.692	± .020[d]
.080	12.390	14.430	4.821	10.601	10.701	± .020[d]
.100	12.434	14.486	4.827	10.600	10.711	± .020[d]

[a] ϵ_a values are given to the third decimal place to preserve the smooth variation of the data with concentration, although the uncertainties are in the second decimal place.

[b] Wavelength 345.0 nm is near one of the two isosbestic points in $HCrO_4^-/Cr_2O_7^=$ spectra. Because it is on the slope of the composite spectrum, reproduction of the ϵ_a values is dependent on wavelength accuracy. Measurements at this wavelength should be made only for verification of the linearity of the absorbance scale.

[c] ϵ_a values are not corrected for the effects of internal multiple reflections within the cuvette, nor have the weights been corrected to vacuum. With these two exceptions, the uncertainties given include all known systematic errors and the 95 percent confidence interval of the mean.

[d] At wavelength 313.0 nm, the uncertainty is reduced to ±0.010.

*The term "apparent" is used because no corrections have been applied to the data for the effects of internal multiple reflections within the cuvette or for buoyancy, i.e., the weights used to express concentrations have not been corrected to vacuum. These combined corrections do not exceed 0.2 percent. The specific absorbances are given in reference 1.

The nomenclature used in this certificate is that recommended by K. D. Mielenz, Anal. Chem. **48, 1093-1094 (1976), which is reproduced in the Appendix of NBS Special Publication 260-54.

Washington, D.C. 20234
June 1, 1977

J. Paul Cali, Chief
Office of Standard Reference Materials

The overall direction and coordination of the technical measurements leading to this certificate were performed under the joint chairmanship of I. L. Barnes and J. R. DeVoe.

The technical and support aspects involved in the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by R. W. Seward.

We wish to acknowledge: The contributions of K. D. Mielenz, NBS Analytical Chemistry Division, for his many valuable discussions and suggestions, and H. H. Ku, NBS Applied Mathematics Division, for his statistical treatment of the data: the cooperation and early support of the National Institute of General Medical Sciences in the research leading to this SRM; and the encouragement of George N. Bowers, Jr., M.D., Hartford Hospital, Hartford, Conn., and Royden N. Rand, Ph.D., Eastman Kodak Co., Research Laboratories, Rochester, N.Y.

PREPARATION AND CERTIFICATION

The details of the preparation and certification of SRM 935 are provided in NBS Special Publication 260-54, Certification and Use of Acidic Potassium Dichromate Solutions as an Ultraviolet Absorbance Standard [2]. This publication should be referred to every time SRM 935 is to be used. Briefly, the transmittances, T, of the solutions prepared from the undried, as received, material were measured with the NBS Institute for Materials Research high-accuracy transmission spectrometer [3].

The ϵ_a values were calculated for each wavelength using the relation:

$$\epsilon_a = \frac{D_s - D_b}{b \times c} = \frac{A_a}{b \times c} \qquad (1)$$

where:

ϵ_a = apparent specific absorbance
A_a = apparent absorbance
D_s = transmittance density of the sample solution, $-\log_{10} T_s$
D_b = transmittance density of the blank solution, $-\log_{10} T_b$
b = internal cuvette pathlength, cm
c = concentration, by weight, of $K_2Cr_2O_7$ solution, $g \cdot kg^{-1}$

The crystalline potassium dichromate used for SRM 935 is a special lot of analytical reagent grade material obtained from the J. T. Baker Chemical Co., Phillipsburg, N.J.

Assay: A coulometric assay of the purity of the undried material was performed by G. Marinenko of the NBS Analytical Chemistry Division. The purity of SRM 935, expressed as an oxidant, was found to be 99.972 ± 0.005 percent where the uncertainty figure represents the 95 percent confidence interval for the mean based on 11 degrees of freedom. In addition, the material was examind by optical emission spectrometry for trace elemental impurities by J. A. Norris of the NBS Analytical Chemistry Division. The only significant impurities detected were sodium and rubidium. Their concentrations were estimated to be in the range of 0.02 and 0.03 percent, respectively. Drying at 105 °C for 12 hours showed that the surface moisture of this material was less than 0.01 percent.

Stability: Solutions prepared from SRM 935 in the concentration range indicated in table 1 and made according to the instructions given in NBS SP 260-54 have been found to be stable within the uncertainties given in table 1 for at least six months when stored at room temperature and protected from evaporation and exposure to light.

INSTRUCTIONS FOR USE

The use of SRM 935 as an absorbance standard requires the careful preparation of a series of solutions of known concentrations, c, of the potassium dichromate in 0.001 N perchloric acid. These solutions are transferred to a quartz cuvette of known pathlength, b, and their apparent absorbances measured at wavelengths 235, 257, 313, and 350 nm, using the spectral bandpass requirements given in table 1. The preparation and measurement of these solutions are described in detail in Section 5 of NBS SP 260-54.

The accuracy of the absorbance scale of the spectrometer being tested is ascertained by comparing the measured apparent absorbances, A_a, of a series of 0.001 N perchloric acid solutions containing 0.020 to 0.100 gram $K_2Cr_2O_7$/kg to the A_a values calculated from the certified ϵ_a values. Although the ϵ_a data in table 1 are given for nominal concentrations of 0.020, 0.040, 0.060, 0.080, and 0.100 g $K_2Cr_2O_7$/kg, the ϵ_a values for concentrations between these nominal concentrations can be determined by linear interpolation. Using the appropriate ϵ_a values, the calculated A_a values at 23.5 °C are obtained from the expression:

$$A_a = \epsilon_a \times b \times c \qquad (2)$$

<u>Calculations:</u>

An example of the calculation of A_a for one concentration of $K_2Cr_2O_7$ under a specified set of conditions is shown below. Calculations of A_a for other concentrations and wavelengths are performed in a similar manner.

Conditions: Wavelength = 350 nm, spectral bandpass 0.8 nm or less
$b = 0.9982$ cm
$c = 0.04375\ g{\cdot}kg^{-1}$
$t = 23.5$ °C

From column 6, table 1, the ϵ_a for concentrations of 0.040 and 0.060 $g{\cdot}kg^{-1}$ are 10.682 and 10.692, respectively. The corresponding ϵ_a for $c = 0.04375\ g{\cdot}kg^{-1}$ is:

$$\epsilon_a = 10.682 + \frac{0.04375 - 0.040}{0.060 - 0.040}(10.692 - 10.682)$$

$$\epsilon_a = 10.682 + 0.0019$$

$$\epsilon_a = 10.684$$

The calculated apparent absorbance, A_a, from equation 2, is:

$$A_a = 10.684 \times 0.9982 \times 0.04375$$

$$A_a = 0.4666$$

The uncertainty, ΔA_a, in the calculated A_a is determined from the combined uncertainties in ϵ_a, b, and c in equation 2, provided no other systematic errors are present. Thus:

$$\Delta A_a = bc\left|\Delta\epsilon_a\right| + \epsilon_a c\left|\Delta b\right| + \epsilon_a b\left|\Delta c\right| \qquad (3)$$

To evaluate ΔA_a, $\Delta\epsilon_a$ is taken from column 7 of table 1 and the Δb and Δc values must be determined experimentally.

In the experiments performed to obtain the ϵ_a values in table 1, the uncertainties for b and c did not exceed 1 part in 10^4 and 2 parts in 10^4, respectively.

The solution of equation 3 gives:

$$\Delta A_a = 1(0.044)\,(0.020) + 10.7(0.044)\,(0.0001) + 10.7(1)\,(0.0000088)$$
$$= 0.0010$$

Thus, the uncertainty of A_a, for the above set of conditions, is ±0.0010.

The correction of the absorbance scale of the absorption spectrometer under test is determined by plotting the differences between A_a measured and A_a calculated as a function of absorbance. A typical plot of such a graph is shown in figure 1. The apparent absorbances measured on this instrument at 350 nm are accurate when the indicated correction is subtracted from the corresponding absorbance scale reading, provided that the conditions of wavelength accuracy, spectral bandpass, and absence of stray light are fulfilled as specified in NBS SP 260-54. Correction curves for wavelengths 235, 257, and 313 nm are obtained in a similar manner.

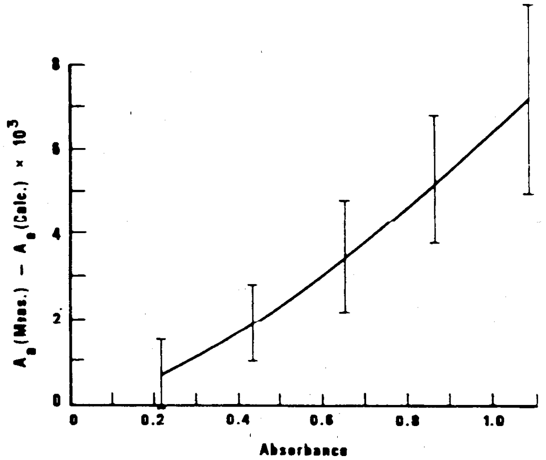


Figure 1. Correction curve for the absorbance scale of a precision spectrometer. The error bars are the sum of the errors arising from the uncertainties in the certified apparent specific absorbances, ϵ_a, cuvette pathlength, b, and concentration, c.

Temperature Correction:

Although ϵ_a values in table 1 are certified at 23.5 °C, SRM 935 can be used as an absorbance standard at other temperatures in the range 20 to 30 °C provided corrections are made to the ϵ_a values. Over this range the apparent specific absorbances decrease linearly with increasing temperature for all the wavelengths given in table 1. The corresponding temperature coefficients, k, for these wavelengths are given in table 2.

Table 2. Variation of ϵ_a with Temperature Over the Range 20 to 30 °C.

λ, nm	Temperature Coefficient, k Percent per degree Celsius
235	−0.05
257	−0.05
313	−0.02
345	−0.08
350	−0.05

The value of ϵ_a at any temperature in the range 20 to 30 °C can be calculated from the certified value and the appropriate temperature coefficient using the relation:

$$\epsilon_a^t = \epsilon_a^{23.5}\left[1 + \frac{k}{100}(t - 23.5)\right]$$

where: ϵ_a^t = apparent specific absorbance at temperature t (°C)

$\epsilon_a^{23.5}$ = apparent specific absorbance certified at 23.5 °C.

k = temperature coefficient, percent per °C.

REFERENCES

1. Burke, R. W., and Mavrodineanu, R., Acidic Potassium Dichromate Solutions as Ultraviolet Absorbance Standards, J. Res. Nat. Bur. Stand. (U.S.), 80A (Phys. and Chem.), No. 4, 631-636 (July-Aug. 1976).

2. Burke, R. W., and Mavrodineanu, R., Certification and Use of Acidic Potassium Dichromate Solutions as an Ultraviolet Absorbance Standard, NBS Spec. Publ. 260-54 (1977). Copies may be obtained from the Office of Standard Reference Materials, National Bureau of Standards, Washington, D.C. 20234.

3. Mavrodineanu, R., An Accurate Spectrophotometer for Measuring the Transmittance of Solid and Liquid Materials, J. Res. Nat. Bur. Stand. (U.S.), 76A (Phys. and Chem.), No. 5, 405-425 (1972).

U.S. Department of Commerce
Juanita M. Kreps
Secretary

National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards

Certificate

Standard Reference Material 2009

Didymium Glass Filter for Checking the Wavelength Scale of Spectrophotometers

Kenneth L. Eckerle and William H. Venable, Jr.

Serial Number:

This SRM is intended for use in calibrating the wavelength scale in the visible wavelength region of scanning spectrophotometers having nominal bandwidths in the range 1.5 to 10.5 nm. Depending upon the bandwidth of the spectrophotometer, anywhere from 14 to 24 wavelength corrections can be determined from 400 to 760 nm. Detailed instructions on the use of this SRM and examples of its use are given in NBS Special Publication 260-66. Each didymium-glass filter is identified by the SRM number and a serial number.

The wavelengths of the transmittance minima as obtained from measurements on two filters representative of the melt are given in Table 1. These values are given for seven equally spaced values of the half-height width of triangular passbands. The minima number is identified in the figure that illustrates the spectral transmittance as a function of wavelength. The wavelength values of nine points of inflection on the spectral transmittance curve as obtained on two filters are given in Table 2. These inflection points are representative of the melt and are also identified in the figure. These inflection points should only be used with the transmittance minima as described in Sections 2.2 and 2.3 in SP 260-66.

The measurements on which these tables are based were made at 25 °C with a high-precision reference spectrophotometer that has a wavelength accuracy of 0.04 nm. Table 3 indicates the estimated random (as obtained from 4 sets of measurements on a single filter) and systematic errors of the transmittance minima given in Table 1. Table 2 also indicates the range of the measured wavelengths of the inflection points. Trial calibrations made on several instruments, using both minima and inflection points, indicate that wavelength corrections made with these SRM's can be accurate to 0.2 nm. The uncertainty of a calibration, however, will depend upon the stability and other characteristics of a particular instrument.

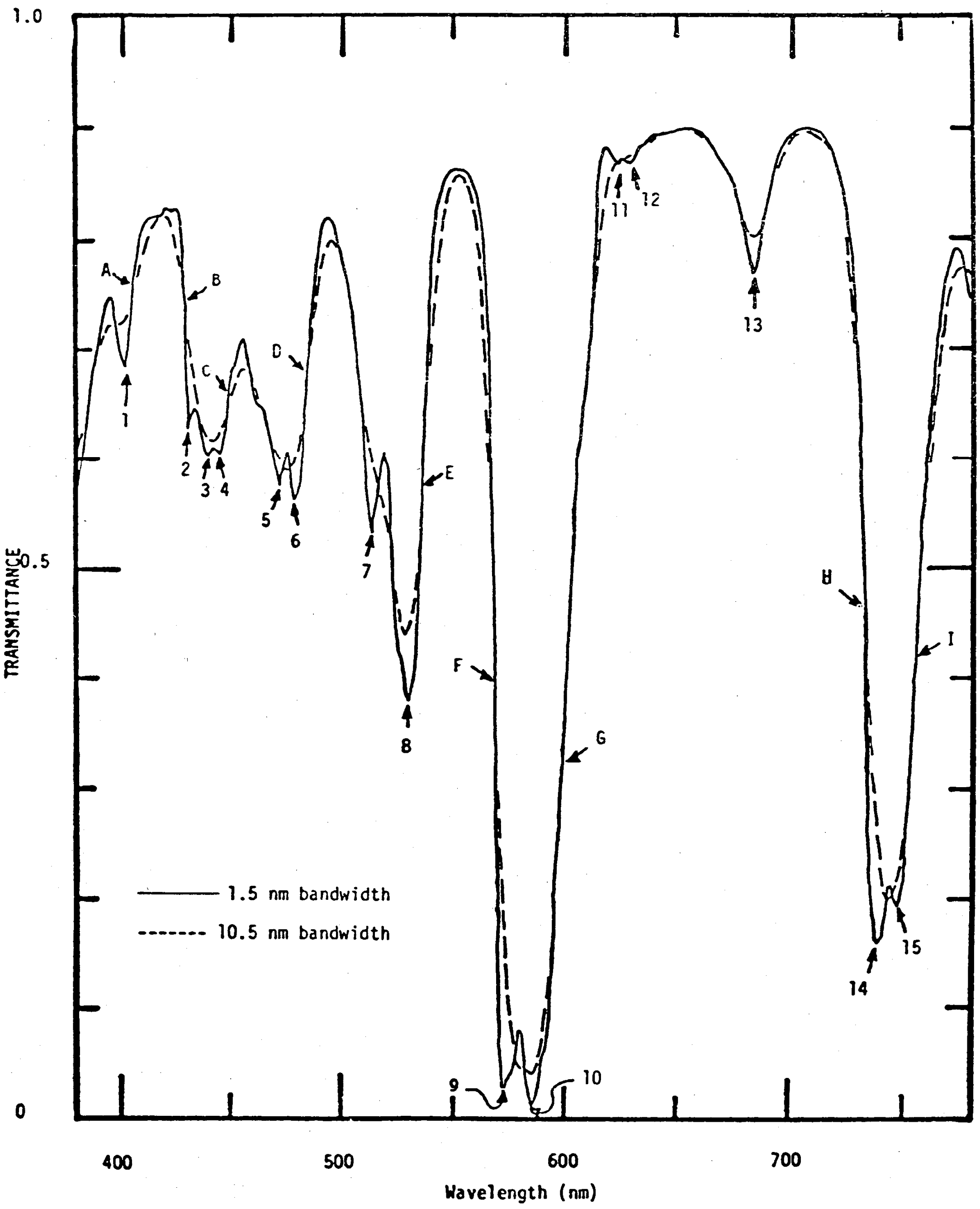
The technical and support aspects involved in the preparation, certification, and issuance of this SRM were coordinated through the Office of Standard Reference Materials by R. K. Kirby.

The spectral transmittance as a function of wavelength for a filter representative of the melt is given in Table 4. These values are not certified but are provided for use as specified in SP 260-66. *They should not be used to check the photometric scale of a spectrophotometer.*

It is recommended that the filter be handled only by its edges and when not in use it should be stored in the box provided. If cleaning is necessary, wet the filter with water and rub gently with optical lens tissue soaked with a mild soap solution, rinse with distilled water, rinse with isoprophyl alcohol, and rinse again with distilled water. Dry after each rinsing by wiping lightly with optical lens tissue.

Washington, D.C. 20234
January 8, 1980

George A. Uriano, Chief
Office of Standard Reference Materials



Spectral transmittance of a typical didymium glass filter. Numbers indicate the principal points of minimum transmittance and letters indicate the principal points of inflection.

Table 1

Certified Wavelengths (nm) of the Transmittance Minima for the Indicated Bandwidths

Bandwidth Minimum No.	1.5 nm	3.0 nm	4.5 nm	6.0 nm	7.5 nm	9.0 nm	10.5 nm
1	402.42	401.81	401.69	401.66	401.42	400.95	
2	431.50	432.48					
3	440.27	440.52	441.84	442.52	442.37	442.08	441.33
4	445.59	445.14					
5	472.72	472.58	472.88				
6	478.89	479.34	479.28	478.31	477.36	476.50	475.65
7	513.45	513.61	513.89	514.31	515.38		
8	529.58	530.02	529.90	529.47	529.27	529.12	528.88
9	572.69	573.27	574.21	575.11	576.59		
10	585.34	585.54	585.77	586.02	585.99	585.35	584.42
11	623.62	624.02					
12	629.53	629.41	628.56	627.03	627.02		
13	684.66	684.68	684.71	684.72	684.71	684.66	684.58
14	739.86	739.96	740.24	740.91	742.01	742.97	743.65
15	748.28	748.10					

Table 2

Wavelengths and Transmittances at Nine Selected Points of Inflection

Point Identification	Wavelength (nm)	Range* (nm)	Transmittance†
A	406.44	+.08 −.06	0.7760
B	429.43	+.05 −.05	.7359
C	449.49	+.06 −.06	.6516
D	484.84	+.10 −.15	.6758
E	536.50	+.06 −.09	.5805
F	568.15	+.08 −.04	.4023
G	599.05	+.05 −.07	.3348
H	733.39	+.06 −.03	.4719
I	756.45	+.01 −.02	.4177

*The range of wavelengths within which the wavelength for the given transmittance will fall for symmetric triangular passbands with half-height bandwidths from 1.5 to 10.5 nm.

†These values of transmittance are not certified.

Table 3

Estimated Random and Systematic Errors of the Transmittance Minima

Band Number	Nominal Wavelength of Minimum Transmittance	Standard Deviation for Indicated Bandwidth						
		1.5 nm	3.0 nm	4.5 nm	6.0 nm	7.5 nm	9.0 nm	10.5 nm
1	402 nm	0.015 nm (0.07)[†]	0.018 nm	0.020 nm	0.021 nm	0.025 nm	0.029 nm	–
2	431	0.013 (0.06)	0.010	–	–	–	–	–
3	440	0.021 (0.05)	0.013	0.050	0.028	0.009	0.005	0.007 nm (0.25)
4	446	0.023 (0.06)	0.029	–	–	–	–	–
5	473	0.011 (0.08)	0.012	0.032	–	–	–	–
6	479	0.015 (0.15)	0.013	0.009	0.009	0.009	0.014	0.017 (0.25)
7	573	0.022 (0.08)	0.016	0.014	0.013	0.010	–	–
8	530	0.012 (0.21)	0.010	0.010	0.011	0.010	0.011	0.010 (0.25)
9	573	0.004 (0.05)	0.010	0.010	0.012	0.014	–	–
10	585	0.007 (0.06)	0.004	0.007	0.008	0.008	0.007	0.007 (0.25)
11	624	0.058 (0.06)	0.061	–	–	–	–	–
12	630	0.210 (0.02)	0.120	0.171	0.133	0.091	–	–
13	685	0.019 (0.05)	0.029	0.024	0.017	0.014	0.014	0.012 (0.25)
14	740	0.009 (0.25)	0.013	0.011	0.010	0.009	0.009	0.010 (0.25)
15	748	0.020 (0.04)	0.016	–	–	–	–	–

[†] Values in parentheses are estimates of the systematic error. The estimates for the 1.5 nm bandwidth were obtained from the data taken at 1.5 nm intervals as compared to data taken at 0.15 nm intervals. The method of estimating the systematic errors for the 10.5 nm bandwidth is described in Section 3.4 in SP 260-66.

TABLE 4

Transmittance (T) as a function of wavelength from 380 to 780.5 nm for a bandwidth of 1.5 nm. The estimates of random and systematic errors are also provided. These data are not certified.

Wave-Length (nm)	T	ΔT Standard Error	ΔT Systematic	Wave-Length (nm)	T	ΔT Standard Error	ΔT Systematic
380.00	.61832	.000054	.00010	381.50	.63896	.000085	.00010
383.00	.66210	.000064	.00010	384.50	.68291	.000079	.00010
386.00	.70112	.000047	.00010	387.50	.71885	.000071	.00010
389.00	.73590	.000095	.00010	390.50	.74954	.000040	.00010
392.00	.76201	.000083	.00010	393.50	.77104	.000055	.00010
395.00	.77620	.000064	.00010	396.50	.77123	.000038	.00010
398.00	.75013	.000069	.00010	399.50	.72419	.000079	.00010
401.00	.70966	.000070	.00010	402.50	.69505	.000071	.00010
404.00	.73683	.000057	.00010	405.50	.75886	.000047	.00010
407.00	.77756	.000088	.00010	408.50	.80673	.000072	.00010
410.00	.82277	.000058	.00010	411.50	.82813	.000038	.00010
413.00	.83115	.000060	.00010	414.50	.83287	.000075	.00010
416.00	.83222	.000060	.00010	417.50	.83200	.000029	.00010
419.00	.83277	.000059	.00010	420.50	.83844	.000066	.00010
422.00	.83711	.000064	.00010	423.50	.83328	.000052	.00010
425.00	.83511	.000049	.00010	426.50	.83414	.000045	.00010
428.00	.81620	.000055	.00010	429.50	.72877	.000023	.00010
431.00	.62212	.000096	.00010	432.50	.63478	.000039	.00010
434.00	.64183	.000057	.00010	435.50	.63392	.000059	.00010
437.00	.61858	.000049	.00010	438.50	.60090	.000038	.00010
440.00	.59163	.000064	.00010	441.50	.59496	.000053	.00010
443.00	.60082	.000063	.00010	444.50	.59576	.000068	.00010
446.00	.59315	.000055	.00010	447.50	.60769	.000044	.00010
449.00	.63551	.000083	.00010	450.50	.66313	.000060	.00010
452.00	.68002	.000069	.00010	453.50	.69110	.000057	.00010
455.00	.70206	.000027	.00010	456.50	.70341	.000033	.00010
458.00	.69051	.000025	.00010	459.50	.66807	.000049	.00010
461.00	.64890	.000057	.00010	462.50	.63966	.000060	.00010
464.00	.63837	.000070	.00010	465.50	.63353	.000040	.00010
467.00	.62094	.000072	.00010	468.50	.60572	.000053	.00010
470.00	.59042	.000008	.00010	471.50	.57213	.000072	.00010
473.00	.56160	.000033	.00010	474.50	.58414	.000031	.00010
476.00	.59528	.000052	.00010	477.50	.56003	.000052	.00010
479.00	.54953	.000070	.00010	480.50	.55659	.000036	.00010
482.00	.57326	.000025	.00010	483.50	.62009	.000040	.00010
485.00	.67764	.000052	.00010	486.50	.72126	.000072	.00010
488.00	.75412	.000045	.00010	489.50	.78321	.000082	.00010
491.00	.80556	.000035	.00010	492.50	.81759	.000060	.00010
494.00	.82117	.000063	.00010	495.50	.81916	.000038	.00010
497.00	.81233	.000096	.00010	498.50	.80115	.000044	.00010

TABLE 4 (cont)

Wave-Length (nm)	T	ΔT Standard Error	ΔT Systematic	Wave-Length (nm)	T	ΔT Standard Error	ΔT Systematic
500.00	.78689	.000060	.00010	501.50	.77050	.000062	.00010
503.00	.75433	.000047	.00010	504.50	.73899	.000046	.00010
506.00	.72185	.000070	.00010	507.50	.69622	.000054	.00010
509.00	.65070	.000060	.00010	510.50	.58889	.000023	.00010
512.00	.53799	.000044	.00010	513.50	.51737	.000050	.00010
515.00	.53523	.000035	.00010	516.50	.56547	.000033	.00010
518.00	.58503	.000042	.00010	519.50	.59603	.000038	.00010
521.00	.57933	.000049	.00010	522.50	.51464	.000058	.00010
524.00	.43678	.000044	.00010	525.50	.40684	.000029	.00010
527.00	.39690	.000057	.00010	528.50	.36734	.000019	.00010
530.00	.36174	.000012	.00010	531.50	.37185	.000046	.00010
533.00	.38958	.000046	.00010	534.50	.45072	.000037	.00010
536.00	.54457	.000052	.00010	537.50	.63283	.000038	.00010
539.00	.69863	.000066	.00010	540.50	.75111	.000028	.00010
542.00	.79247	.000062	.00010	543.50	.82093	.000050	.00010
545.00	.84006	.000043	.00010	546.50	.85304	.000068	.00010
548.00	.86137	.000073	.00010	549.50	.86532	.000043	.00010
551.00	.86587	.000068	.00010	552.50	.86526	.000035	.00010
554.00	.86482	.000057	.00010	555.50	.86353	.000038	.00010
557.00	.85997	.000035	.00010	558.50	.85316	.000088	.00010
560.00	.84237	.000045	.00010	561.50	.82504	.000060	.00010
563.00	.79728	.000048	.00010	564.50	.75134	.000031	.00010
566.00	.66681	.000052	.00010	567.50	.50244	.000030	.00010
569.00	.25719	.000033	.00010	570.50	.07751	.000011	.00010
572.00	.02604	.000010	.00010	573.50	.02536	.000013	.00010
575.00	.03443	.000011	.00010	576.50	.03857	.000013	.00010
578.00	.05117	.000012	.00010	579.50	.06944	.000014	.00010
581.00	.06863	.000007	.00010	582.50	.03836	.000014	.00010
584.00	.01549	.000007	.00010	585.50	.01027	.000006	.00010
587.00	.01577	.000016	.00010	588.50	.02979	.000022	.00010
590.00	.04702	.000023	.00010	591.50	.05748	.000009	.00010
593.00	.07588	.000013	.00010	594.50	.11929	.000028	.00010
596.00	.19036	.000034	.00010	597.50	.26661	.000020	.00010
599.00	.32378	.000038	.00010	600.50	.38146	.000043	.00010
602.00	.44667	.000041	.00010	603.50	.51047	.000043	.00010
605.00	.56569	.000048	.00010	606.50	.60327	.000074	.00010
608.00	.62507	.000047	.00010	609.50	.64527	.000041	.00010
611.00	.68076	.000072	.00010	612.50	.73522	.000054	.00010
614.00	.79774	.000073	.00010	615.50	.84664	.000067	.00010
617.00	.87427	.000086	.00010	618.50	.88467	.000056	.00010

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TABLE 4 (cont)

Wave-Length (nm)	T	ΔT Standard Error	ΔT Systematic	Wave-Length (nm)	T	ΔT Standard Error	ΔT Systematic
620.00	.88413	.000042	.00010	621.50	.87711	.000071	.00010
623.00	.87047	.000037	.00010	624.50	.87083	.000060	.00010
626.00	.87359	.000066	.00010	627.50	.87310	.000069	.00010
629.00	.87008	.000069	.00010	630.50	.87085	.000064	.00010
632.00	.87569	.000048	.00010	633.50	.88221	.000043	.00010
635.00	.88828	.000124	.00010	636.50	.89117	.000089	.00010
638.00	.89322	.000035	.00010	639.50	.89543	.000062	.00010
641.00	.89755	.000085	.00010	642.50	.89905	.000051	.00010
644.00	.90014	.000052	.00010	645.50	.90080	.000043	.00010
647.00	.90124	.000041	.00010	648.50	.90112	.000060	.00010
650.00	.90106	.000034	.00010	651.50	.90149	.000079	.00010
653.00	.90218	.000054	.00010	654.50	.90300	.000066	.00010
656.00	.90301	.000082	.00010	657.50	.90256	.000044	.00010
659.00	.90198	.000049	.00010	660.50	.90115	.000091	.00010
662.00	.89951	.000104	.00010	663.50	.89656	.000071	.00010
665.00	.89155	.000045	.00010	666.50	.88540	.000079	.00010
668.00	.87844	.000079	.00010	669.50	.87177	.000085	.00010
671.00	.86691	.000077	.00010	672.50	.86284	.000056	.00010
674.00	.85883	.000035	.00010	675.50	.85369	.000076	.00010
677.00	.84419	.000077	.00010	678.50	.83115	.000053	.00010
680.00	.81611	.000076	.00010	681.50	.79779	.000063	.00010
683.00	.77773	.000111	.00010	684.50	.76323	.000051	.00010
686.00	77287	.000111	.00010	687.50	.79255	.000057	.00010
689.00	.80954	.000080	.00010	690.50	.82685	.000045	.00010
692.00	.84399	.000066	.00010	693.50	.85936	.000052	.00010
695.00	.87138	.000058	.00010	696.50	.88070	.000036	.00010
698.00	.88764	.000054	.00010	699.50	.89254	.000029	.00010
701.00	.89577	.000056	.00010	702.50	.89798	.000040	.00010
704.00	.89957	.000049	.00010	705.50	.90089	.000060	.00010
707.00	.90166	.000056	.00010	708.50	.90231	.000071	.00010
710.00	.90225	.000066	.00010	711.50	.90168	.000071	.00010
713.00	.90041	.000056	.00010	714.50	.89850	.000060	.00010
716.00	.89593	.000042	.00010	717.50	.89232	.000040	.00010
719.00	.88755	.000075	.00010	720.50	.88137	.000049	.00010
722.00	.87336	.000061	.00010	723.50	.86256	.000086	.00010
725.00	.84758	.000054	.00010	726.50	.82620	.000070	.00010
728.00	.79437	.000071	.00010	729.50	.74399	.000039	.00010
731.00	.66643	.000039	.00010	732.50	.55165	.000033	.00010
734.00	.40413	.000053	.00010	735.50	.26498	.000040	.00010
737.00	.17772	.000064	.00010	738.50	.14784	.000028	.00010

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TABLE 4 (cont)

Wave-Length (nm)	T	ΔT Standard Error	ΔT System-atic	Wave-Length (nm)	T	ΔT Standard Error	ΔT System-atic
740.00	.14400	.000017	.00010	741.50	.14841	.000015	.00010
743.00	.16928	.000022	.00010	744.50	.19331	.000020	.00010
746.00	.19302	.000027	.00010	747.50	.17683	.000014	.00010
749.00	.17678	.000040	.00010	750.50	.20220	.000026	.00010
752.00	.24335	.000050	.00010	753.50	.29149	.000049	.00010
755.00	.34682	.000095	.00010	756.50	.40869	.000063	.00010
758.00	.47017	.000099	.00010	759.50	.52523	.000164	.00010
761.00	.57256	.000071	.00010	762.50	.61237	.000077	.00010
764.00	.64917	.000089	.00010	765.50	.68412	.000093	.00010
767.00	.71596	.000088	.00010	768.50	.74294	.000080	.00010
770.00	.76342	.000040	.00010	771.50	.77754	.000144	.00010
773.00	.78603	.000082	.00010	774.50	.78934	.000082	.00010
776.00	.78665	.000098	.00010	777.50	.77668	.000050	.00010
779.00	.75800	.000083	.00010	780.50	.73040	.000120	.00010

SRM 2009

U.S. Department of Commerce
Elliot L. Richardson,
Secretary

National Bureau of Standards
Ernest Ambler, Acting Director

National Bureau of Standards
Certificate
Standard Reference Material 2030
Glass Filter for Transmittance Measurement

R. Mavrodineanu and J. R. Baldwin

This Standard Reference Material is intended as a reference source for one-point verification of the transmittance and absorbance scales of spectrophotometers at the given wavelength and measured transmittance. It consists of one glass filter in its holder and one empty filter holder. The filter bears an identification number. For protection, the metal holder is provided with two shutters that should be removed during measurements.

The transmittance, T, at λ 465.0 nm and for a spectral bandpass of 2.7 nm is: ________________. The corresponding transmission density, $-\log_{10}T$, is: ________________.

Date of Certification: ________________ ________________.

The transmittance value (T) can be converted to percent transmittance (%T) by multiplying by 100. The transmission density is calculated from the measured transmittance (T). This value should be indicated by the absorbance scale of the spectrophotometer if the filter is measured against air.

The transmittance value given was measured against air at an ambient temperature of 23.5 °C, and is estimated to be accurate to within ±0.5 percent at the time of certification. This uncertainty includes the random and systematic errors of the calibration procedure, as well as transmittance changes of the filter during the period of calibration.

It is possible that aging of the glass may cause some filters to change transmittance by about ±1 percent over a period of approximately 1 year from the date of calibration. Improper storage or handling of the filter may also cause changes [5]. In cases where verification is desirable, the filter should be returned to the National Bureau of Standards for cleaning and recalibration.

It is recommended that the filter in the holder be handled only by the edges and with soft plastic (polyethylene) gloves and optical lens tissue. When not in use, it should be stored in its holder, with the shutters on, and in the box provided for this purpose. Extended exposure to laboratory atmosphere and dusty surroundings should be avoided.

This Standard Reference Material was issued with the assistance and advice of K. D. Mielenz and J. R. DeVoe.

The technical and support aspect involved in the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by R. W. Seward.

Washington, D.C. 20234
September 23, 1976

J. Paul Cali, Chief
Office of Standard Reference Materials

The transmittance measurements were made using the high-accuracy spectrophotometer designed and built at the Institute for Materials Research, National Bureau of Standards [1]. This instrument represents the primary transmittance standard; its transmittance accuracy was established using the double-aperture method of linearity testing [1,3,5,6]. The reproducibility (relative standard deviation obtained from a set of 20 measurements) of this high-accuracy spectrometer for transmittance measurements on glass filters similar to the one used to produce this SRM, is approximately 0.02 percent.

The combined uncorrected systematic error of the instrument is estimated to be not more than $\pm 10^{-4}$ transmittance units.

As indicated on the face of this certificate, SRM 2030 is intended to be used as a one-point verification of the transmittance (absorbance) scales of spectrophotometers. If these scales require verification at other wavelengths and transmittances, "Glass Filters for Spectrophotometry," SRM 930c or subsequent issues, should be used.

The neutral glass for the filter was provided by Schott of Mainz, Germany, and is designated as "Jena Color and Filter Glass " [2,5].

The exposed surface of the glass is approximately 29 × 8 mm, measuring from a point 1.5 mm above the base of the filter holder (see figure). The empty filter holder is provided to be used in the reference beam of the spectrophotometer so that approximately equivalent conditions of stray radiations are achieved for both beams.

The transmittance of the filter depends upon the intrinsic properties of the material, spectral bandpass, wavelength, geometry of the optical beam, temperature, and positioning of the filter. A change of ambient temperature of 1 or 2 °C from 23.5 °C will not significantly affect the calibration [5].Changes in the transmittance may be caused by surface conditions, aging of the glass, exposure to a harmful atmosphere, or careless handling as indicated on the face of this certificate [2,3,4,5]. The effective spectral bandpass used to determine the certified value is given on the face of the certificate and the transmittance measurements are made by producing the vertical image of the slit (about 8 mm by 0.5 mm), using a convergent beam geometry with an aperture ratio f:10, in the middle of the entrance face of the glass filter. The filter was measured in the spectrophotometer in a position perpendicular to the incident light beam as shown in the attached figure.

Prior to certification measurements, the filter was examined for surface defects and thoroughly cleaned [5]. Should the surface of the glass filter become contaminated, no attempt should be made to clean it unless the user has the facilities to demonstrate that the cleaning treatment has not altered the glass surface or degraded the accuracy of the certified value. Because the Standard Reference Material is a transfer standard, the only means available to verify its integrity is remeasuring its transmittance with a primary standard instrument similar to that used in this certification [1,4,5]. In most cases where verification, or recertification, of the transmittance value is desirable, it will be most expeditious to return the filter to the National Bureau of Standards for measurement. Prior to shipment the NBS Office of Standard Reference Materials should be contacted regarding the conditions and cost of verification and/or recertification.

REFERENCES

[1] R. Mavrodineanu, An Accurate Spectrophotometer for Measuring the Transmittance of Solid and Liquid Materials, NBS Journal of Research **76A**, No. 5, 405-425 (1972).

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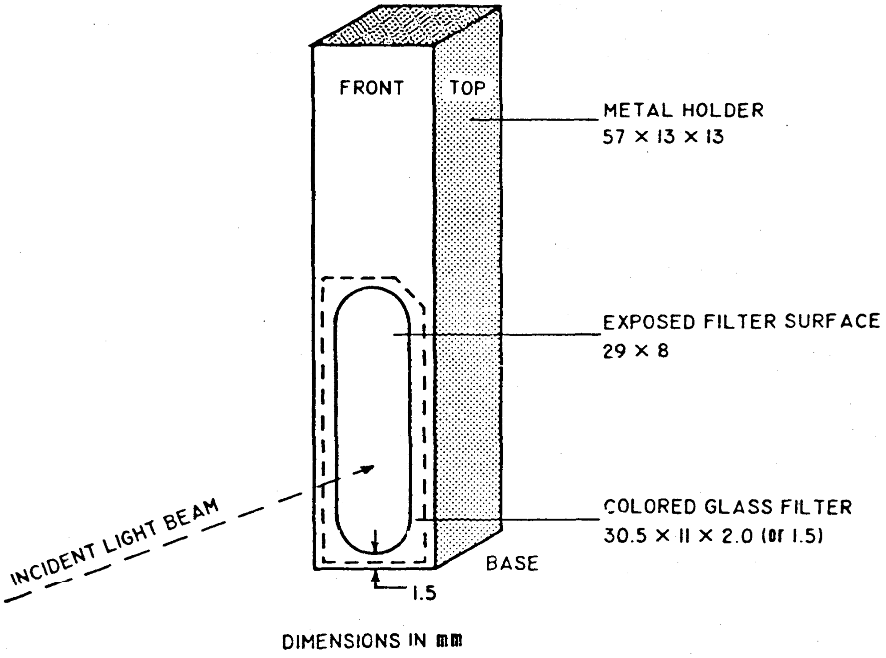
[3] K. S. Gibson, Spectrophotometry, NBS Circ. 484 (Sept. 1949).

[4] Collected Papers from NBS Conference on Accuracy in Spectrophotometry and Luminescence Measurements, NBS Journal of Research **76A**, No. 5, 375-510 (1972).

[5] R. Mavrodineanu and J. R. Baldwin, Glass Filters as a Standard Reference Material for Spectrophotometry-Selection, Preparation, Certification, Use SRM 930, NBS Special Publication 260-51, U.S. Government Printing Office, Washington, D.C. 20402, (1975).

[6] K. D. Mielenz and K. L. Eckerle, Spectrophotometric Linearity Testing Using Double-Beam-Aperture Method, Appl. Optics, **11**, 2294-2303 (1972).

METAL HOLDER FOR THE COLORED GLASS FILTERS



U. S. Department of Commerce
Malcolm Baldrige
Secretary

National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards

Certificate of Calibration

Standard Reference Material 2031

Metal-on-Quartz Filters for Spectrophotometry

This Standard Reference Material is intended for use in the verification of the transmittance and absorbance scales of conventional spectrophotometers in the ultraviolet and visible regions of the electromagnetic spectrum. It consists of three individual filters in their metal holders and one empty filter holder. Two filters, having nominal transmittances of 10 and 30 percent, were produced by evaporating different thicknesses of chromium metal on 1.5-mm thick fused silica plates that had been precision ground and polished. These metal films are protected by 1.5-mm clear fused silica cover plates optically contacted to the base plates. The third filter is a single fused silica plate 3-mm thick, having a nominal transmittance of 90 percent. The metal holders for these filters are provided with shutters to protect the filters when not in use. The shutters must be removed at the time of measurement and be replaced after the measurements have been completed. Each filter holder bears the set and filter identification number.

Set and Filter Ident. Number	TRANSMITTANCE (T)									
	Wavelength, nm									
	250.0	280.0	340.0	360.0	400.0	465.0	500.0	546.1	590.0	635.0

Set and Filter Ident. Number	TRANSMITTANCE DENSITY ($-\log_{10}T$)									
	Wavelength, nm									
	250.0	280.0	340.0	360.0	400.0	465.0	500.0	546.1	590.0	635.0

Date of Certification:

Washington, D.C. 20234
September 20, 1982
(Revision of Certificate
dated 6-1-79)

George A. Uriano, Chief
Office of Standard Reference Materials

The transmittance values (T) can be converted to percent transmittance (%T) by multiplying by 100. The transmittance densities were calculated from the measured transmittance (T). These transmittance densities should be indicated by the absorbance scale of the spectrophotometer when the filters are measured against air. All of the certified transmittance values were obtained by measuring against air at an ambient temperature of 23.5 °C.

The uncertainties of the certified transmittance values of these filters do not exceed 1.0 percent. These uncertainties include 0.5 percent for random errors of the calibration procedure, as well as 0.5 percent for possible changes in the transmittance with time. The long-term stability of the filters with the chromium film has not been rigorously established. Measurements to date, however, suggest that the transmittance of the chromium-coated filters could change by as much as 0.5 percent in the first year after calibration. Therefore, this SRM should be returned to the National Bureau of Standards for free verification of the transmittance values on the first anniversary of the certification date. Information regarding subsequent verifications or recalibrations, including costs for such service, will be included with this first verification report. Before returning this SRM to NBS, information regarding such shipment should be obtained from the Office of Standard Reference Materials, Room B311, Chemistry Building, National Bureau of Standards, Washington, DC 20234. Telephone: (301) 921-2641.

When not in use, the filters should be stored in their holders with the shutters in place and in the metal container provided for this purpose. Extended exposure to laboratory atmosphere and dirty surroundings should be avoided.

The transmittance measurements were made using the high-accuracy spectrophotometer designed and built in the NBS Center for Analytical Chemistry [*1*]. This instrument is the primary transmittance standard; its transmittance accuracy was established using the double-aperture method of linearity testing [1,3,5,6].

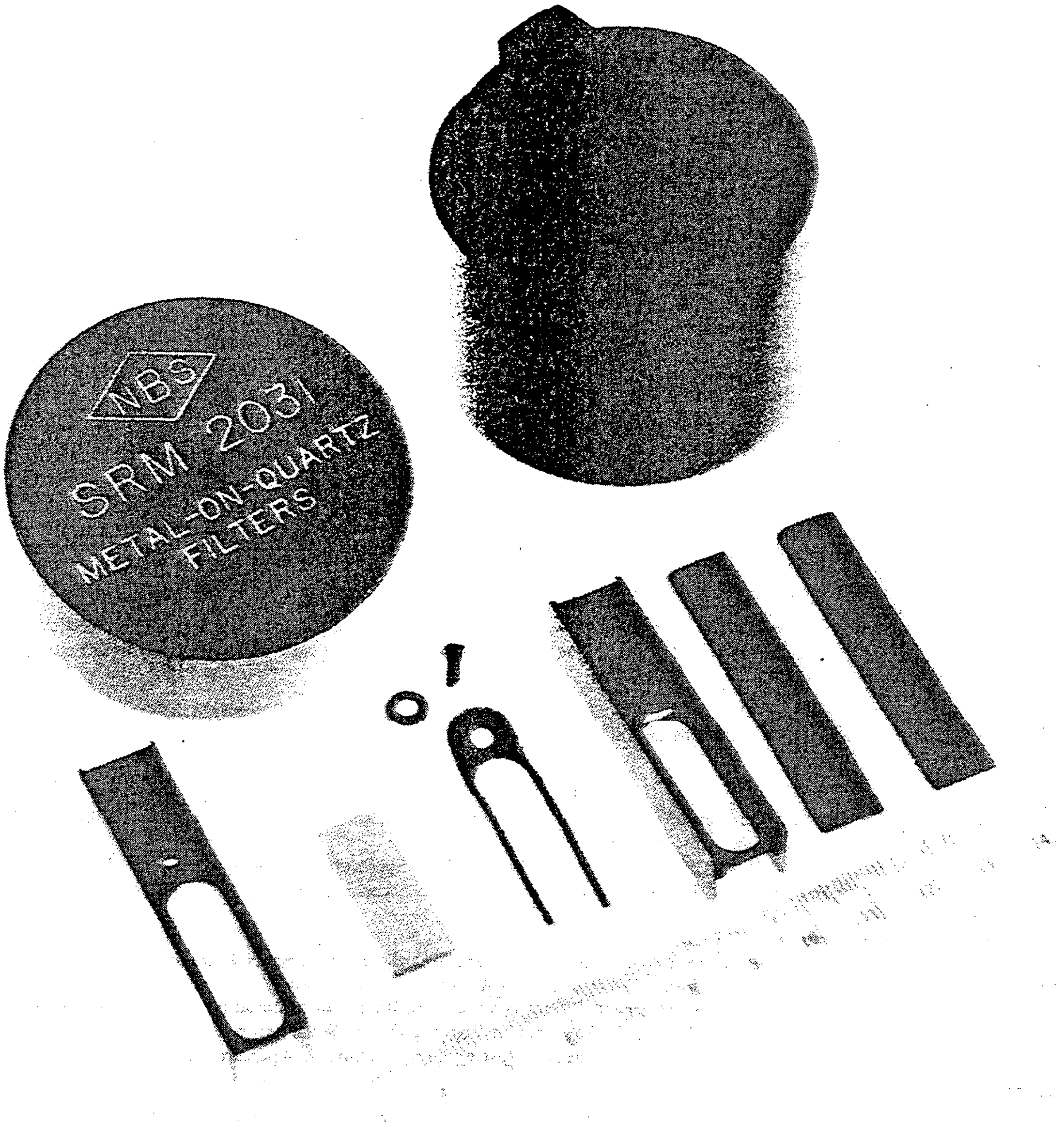
Transmittance measurements for SRM 2031 are made by producing the vertical image of the slit (about 8 mm by 1.5 mm), using a convergent beam geometry with an aperture ratio f:10, in the middle of the entrance face of the filter. The filters were measured in the spectrophotometer in a position perpendicular to the incident light beam. A spectral bandpass of 1.6 nm was used for measurements at all wavelengths. Because the transmittances of these filters exhibit an appreciable optical neutrality, the dependence of transmittance on bandpass is not critical and wider bandpasses may be used in routine measurements. For a quantitative discussion of this subject, the user should consult reference 5, pp. 32 and 33.

The transmittance, T, was measured against air in the reference beam; hence it includes the reflection losses that occur at the air-filter interface. Under these circumstances the measured transmittance, T, has a corresponding transmittance density, defined as $-\log_{10} T$. The internal transmittance, T_i, of a material is defined as the transmittance of the material corrected for reflection losses. This is obtained experimentally when the measurements are made against a blank sample in the reference beam. The absorbance, A, of a material is related to this internal transmittance, T_i, by the expression, $A = \log_{10} T_i$.

The exposed surface of each filter is approximately 29 x 8 mm, measuring from a point 1.5 mm above the base of the filter holder (see figure). The empty filter holder provided is to be used in the reference beam of the spectrophotometer so that approximately equivalent conditions of stray radiation are achieved for both beams. The transmittance of the filters depends upon the intrinsic properties of the material, wavelength, spectral bandpass, geometry of the optical beam, temperature, and positioning of the filter. While changes in ambient temperature of 1 or 2 °C from 23.5 °C have not significantly affected the calibration, the effect of temperature variations exceeding 2 °C have not been investigated. Changes in the transmittance may be caused by surface conditions, aging of the material, exposure to a harmful atmosphere, or careless handling [2,3,4,5].

SRM 2031 is stored in a black anodized aluminum container provided with a threaded cap made of the same metal. Each filter is placed in a cylindrical cavity to prevent any contact between the filter face and the walls of the storage container. Contamination of the filter surface with particulate matter due to static charges is minimized through the metallic nature of the container. A flat leaf spring is inserted into the cylindrical cavity with each filter holder to minimize damage during transportation. These springs can be removed during normal use in the laboratory.

The filter is shown in the assembled unit with its front surface facing up. The filter, in its filter holder, should be placed in the cuvette compartment of the spectrophotometer with its front surface facing the incident light beam and the rear surface facing the photodetector.



Top: Cylindrical container with its screw cap, both made of black-anodized aluminum alloy. Four filter holders can be stored in the cylindrical container. Bottom (from left to right): Aluminum alloy filter holder, 12.5 mm square and 58 mm high; Metal-on-quartz filter; Retaining spring of beryllium-copper with nylon screw and washer; Assembled unit; and Two Delrin shutters. All metal and plastic parts are flat black.

Prior to certification measurements, each filter was examined for surface defects and the condition of the optical contact [5]. Should the surface of the filter become contaminated, no attempt should be made to clean it unless the user has the facilities to demonstrate that the cleaning treatment will not alter the surface or degrade the accuracy of the certified values. As SRM 2031 is a transfer standard, the only means available to verify its integrity is to remeasure its transmittance with a primary standard instrument similar to that used in this certification [1,5]. In most cases, where verification or recertification of the transmittance values is desirable, it will be most expeditious to return the filters to the National Bureau of Standards for measurement.

Further information concerning the selection, preparation, and properties of SRM 2031 will be found in reference 5.

The research, development, and initial production of this SRM were conducted by R. Mavrodineanu and J.R. Baldwin, NBS Inorganic Analytical Research Division.

The transmittance measurements were performed by R.W. Burke, Inorganic Analytical Research Division, and M.V. Smith, Office of Standard Reference Materials. Technical leadership for the preparation and measurements leading to certification was provided by R.W. Burke.

The overall direction and coordination of technical measurements leading to certification were performed under the chairmanship of E.L. Garner.

The technical and support aspects involved in the preparation, certification, and issuance of this Standard Reference Material were coordinated through the Office of Standard Reference Materials by R.W. Seward.

We wish to acknowledge the cooperation of George N. Bowers, Jr., M.D., of Hartford Hospital, Hartford, Connecticut; Royden N. Rand, Ph.D., of the Eastman Kodak Co. Research Laboratories, Rochester, New York; and Donald S. Young, M.D., Ph.D., of the Mayo Clinic, Rochester, Minnesota.

The contributions of H.E. Bennett and J.M. Bennett of the Michelson Laboratory, Physical Optics Branch, Naval Weapons Center, China Lake, California, who helped in the initial production of SRM 2031 are also gratefully acknowledged.

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6. K.D. Mielenz and K.L. Eckerle, Spectrophotometer Linearity Testing Using the Double-Aperture Method, Appl. Optics 11, 2294-2303 (1972).

U.S. Department of Commerce
Juanita M. Kreps
Secretary

National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards

Certificate

Standard Reference Material 2032

Crystalline Potassium Iodide

Heterochromatic Stray Radiant Energy Standard for Ultraviolet Absorption Spectrophotometry

K. D. Mielenz and R. Mavrodineanu

This Standard Reference Material consists of reagent-grade, crystalline potassium iodide (KI) to be used to assess heterochromatic stray radiant energy (stray light) in ultraviolet absorption spectrophotometers in the spectral region below 260 nm. Stray light is assessed by measuring the spectral absorbance of aqueous solutions of SRM 2032 of known pathlengths and concentrations, and comparing the result with the certified values of the specific absorbance, ϵ.

Specific Absorbance[a]

ϵ (L g^{-1} cm^{-1}), vs Wavelength, λ (nm), at 23.5 °C

λ	240	245	250	255	260	265	270	275
ϵ	25.6	10.67	3.66	1.05	0.260	0.0560	0.0121	0.0031

[a] "Specific absorbance" is defined here as absorbance per unit pathlength and unit concentration. The term "absorptivity" has been avoided since it is ambiguously defined. See K. D. Mielenz. Anal. Chem. 48. 1093-1094 (1976).

The estimated uncertainty of these values is ± 5%, which includes the random and systematic errors of the calibration procedure, as well as variations due to possible instability of the KI. (See, Certification Procedure.)

The material used to produce SRM 2032 was obtained from the J. T. Baker Chemical Co., Phillipsburg, N.J., in two bottles, as reagent-grade (99.8% purity) potassium iodide.

SRM 2032 was issued with the technical assistance of J. R. Baldwin, R. W. Burke, A. L. Cummings, B. I. Diamondstone, and G. A. Sleater, and under the overall direction of I. L. Barnes.

The technical and support aspects concerning preparation, certification, and issuance of this SRM were coordinated through the Office of Standard Reference Materials by R. W. Seward.

Washington, D.C. 20234
October 31, 1979

George A. Uriano, Chief
Office of Standard Reference Materials

Supplementary Information

Material Testing:

Material homogeneity was tested by measuring the absorbance of 1% aqueous solutions of the KI sampled from the top, middle, and bottom of each bottle. These measurements, at 265, 267, and 270 nm, showed no evidence of inhomogeneity. Tests for moisture content (Karl Fischer method) gave an average of 0.007%. This value was sufficiently small that all certified data are based on weighed samples of the undried, as received, material.

Stability of the material to UV and visible light was tested by exposing the KI salt in a low-actinic glass bottle to radiation from a 3 kW xenon-arc lamp at a distance of 25 cm for four days at room temperature. The average absorbances at five wavelengths between 240 and 270 nm of aqueous solutions of the exposed material were 1.1% lower than those of solutions of the unexposed material. In view of the severity of this test, this change was considered minor. Nonetheless, the 1.1% was included in the overall uncertainty of the certified values.

Certification Procedure:

The certified specific absorbances were measured in the NBS Center for Analytical Chemistry high-accuracy spectrophotometer [1, 2], equipped with a deuterium lamp and UV averaging sphere [3, 4]. Thirteen aqueous solutions of KI with concentrations ranging from 0.03 to 30 g L^{-1} were prepared and measured against distilled water, using standard fused-silica cuvettes with pathlengths between 10.001 and 10.009 mm[b]. The absorbance measurements were performed at the temperature, $t = 23.5 \pm 0.5$ °C. A 0.2 nm bandpass was used, and wavelength settings were made with an accuracy of 0.05 nm. The measurements were made on at least three concentrations at each wavelength. These concentrations were chosen so that most absorbance readings fell between 0.1 and 1.0 to minimize stray light and bandwidth errors. The data were found to obey Beer's law.

Thus, the equation,

$$\epsilon = A/c\,\ell \tag{1}$$

was used to calculate the specific absorbances, ϵ, from the measured absorbances, A, and the known values of concentration, c, and pathlength, ℓ .

Limits to random error (two standard deviations) of these measurements (including the photometric imprecision of the spectrophotometer, errors in the repositioning of cuvettes, concentration errors, pathlength uncertainties, temperature errors, Beer's law uncertainties, and bandwidth errors) were determined to be ± 2.4%. The error due to the 0.05 nm uncertainty of the wavelength calibration of the spectrophotometer is ± 1.1%.

The uncertainty of 5% stated on the face of this certificate represents the sum of these errors and the above mentioned 1.1% uncertainty due to instability of the KI. No corrections were applied for the effects of internal reflections inside the sample and reference cuvettes, nor were the weights corrected to vacuum. These and all other sources of error were considered negligible.

The temperature coefficient at 260 nm was determined to be

$$\frac{1}{\epsilon}\,\frac{d\epsilon}{dt} = 0.031\ ^{\circ}C^{-1}. \tag{2}$$

For precision measurements, it is recommended that SRM 2032 be used in thermostated cuvettes at 23.5 °C, or that a temperature correction according to Eq. (2) be applied.

[b] Issued by NBS as SRM 932, Quartz Cuvettes for Spectrophotometry.

Instructions for Use

Storage and Preparation:

SRM 2032 should be stored in the original, low-actinic glass bottle and the cardboard container in which it was issued to protect it from unnecessary exposure to light and humidity. When so stored, the expected stability of this material is at least three years. Thus, until additional stability data are obtained, this material should be used within three years of the date of purchase.

All solutions prepared from SRM 2032 should be made in borosilicate glass containers using distilled water and transfer pipettes (Pasteur type) of the same glass and fitted with rubber bulbs[c]. Use clean spectrophotometer cuvettes, free of scratches, made of non-fluorescent fused silica, and fitted with ground-glass or Teflon stoppers to minimize evaporation; or preferably use NBS SRM 932. Mark all cuvettes to assure the same orientation in the spectrophotometer, and place them into their respective holders. Using transfer pipettes of the type mentioned, rinse each cuvette several times with distilled water. Prepare a solution of KI in distilled water (e.g., 1%), fill the sample cuvette with the KI solution and the reference cuvette with distilled water. Measure the absorbance, leaving the cuvettes in their holders, empty them (using the pipettes), and repeat the rinsing and filling operations until constant absorbance readings are obtained. Fresh solutions should be made before every test.

Measurements:

A 1% KI solution ($c = 10\ \text{g L}^{-1}$) with a 1 cm pathlength exhibits a sharp cutoff in transmittance near 260 nm; i.e., it transmits more than 90% above 273 nm, but less than 0.01% below 258 nm. Therefore, with the monochromator set for a wavelength below 260 nm, any appreciable amount of light detected is heterochromatic stray light, which consists of wavelengths above the cutoff. The amount of stray light in the spectrophotometer at wavelength λ may be determined from the equations:

$$T'(\lambda) = \frac{T(\lambda) + x(\lambda)}{1 + x(\lambda)} \tag{3}$$

$$x(\lambda) = \frac{T'(\lambda) - T(\lambda)}{1 - T'(\lambda)} \tag{4}$$

where:

$x(\lambda)$ is the stray light ratio,

$T'(\lambda)$ is the apparent transmittance,

$T(\lambda)$ is the true transmittance.

The stray light ratio, $x(\lambda)$, is the proportion of heterochromatic stray radiant energy in the spectrophotometer for the wavelength setting λ. Equation (4) forms the theoretical basis for the determination of the stray light ratio by comparing apparent transmittance to true transmittance. For SRM 2032, the true transmittance, $T(\lambda)$, of KI solutions may be calculated as:

$$T(\lambda) = 10^{-\epsilon(\lambda) \cdot c\ell}$$

where $\epsilon(\lambda)$ is the certified specific absorbance given on the face of this certificate.

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[c] Soft glass containers and pipettes contain residual amounts of UV absorbing material, but can be used after proper cleaning. Several rinses, first with isopropyl alcohol and then with distilled water, are generally adequate.

The apparent transmittances of KI solutions depend not only on the stray light ratio of the spectrophotometer, but also on the wavelength accuracy and the spectral bandwidth, which can significantly affect the results obtained because of the steep slope of the absorbance cutoff of KI. A low-pressure mercury discharge lamp is suggested for verifying the wavelength scale, and a bandwidth as narrow as compatible with adequate signal-to-noise ratios should be used.

Many instruments do not permit the direct measurement of transmittances below certain limits, e.g., 1% or 0.1%. For such instruments, the use of SRM 2032 will only measure stray light ratios above these limits. To measure stray light ratios below these limits, the reference beam of the spectrophotometer must be attenuated to extend the transmittance scale into the low-transmittance region.

References

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3. K. D. Mielenz, R. Mavrodineanu, and E. D. Cehelnik, J. Res. NBS **78A**, 631-635 (1974).
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U.S. Department of Commerce
Philip M. Klutznick
Secretary

National Bureau of Standards
Ernest Ambler, Director

National Bureau of Standards

Certificate

Standard Reference Material 2033

Crystalline Potassium Iodide with Attenuator Heterochromatic and Isochromatic Stray Radiant Energy Standard for Ultraviolet Absorption Spectrophotometry

K. D. Mielenz and R. Mavrodineanu

This Standard Reference Material consists of reagent-grade crystalline potassium iodide (KI) and a radiation attenuator to be used to assess the heterochromatic stray radiant energy (stray light) in ultraviolet absorption spectrophotometers in the spectral region below 260 nm.. The radiation attenuator consists of two semitransparent evaporated metal-on-fused silica (non-fluorescent) filters, each having a nominal transmittance of 10%. One filter is mounted in a cuvette-style holder that can be inserted into the sample compartment of the spectrophotometer. The other is mounted in one of the two shutters of the holder. The use of this attenuator permits the expansion of the transmittance scale into the low-transmittance region by providing a two-step attenuation of the reference beam of the spectrophotometer to about 1%. The attenuator can also be used to assess isochromatic stray light by following the instructions given in this certificate.

The heterochromatic stray light is assessed by measuring the spectral absorbance of aqueous solutions of SRM 2033 of known pathlengths and concentrations, and comparing the results with the certified values of the specific absorbance, ϵ.

Specific Absorbance[a]

	ϵ (L g^{-1} cm^{-1}), vs Wavelength, λ (nm), at 23.5 °C							
λ	240	245	250	255	260	265	270	275
ϵ	25.6	10.67	3.66	1.05	0.260	0.0560	0.0121	0.0031

[a] "Specific Absorbance" is defined here as absorbance per unit pathlength and unit concentration. The term "absorptivity" has been avoided since it is ambiguously defined. See K. D. Mielenz, Anal. Chem. 48, 1093-1094 (1976).

The estimated uncertainty of these values is ± 5%, which includes the random and systematic errors of the calibration procedure, as well as variations due to possible instability of the KI. (See Certification Procedure.)

The material used to produce SRM 2033 was obtained from the J.T. Baker Chemical Co., Phillipsburg, N.J., in two bottles, as reagent-grade (99.8% purity) potassium iodide.

The transmittance of the filter mounted in the holder at λ255 nm is ____________,
the transmittance of both filters at λ255 is ____________.

SRM 2033 was issued with the technical assistance of J. R. Baldwin, R. W. Burke, A. L. Cummings, B. I. Diamondstone, and G. A. Sleater, and under the overall direction of E. L. Garner, NBS Inorganic Analytical Research Division.

The technical and support aspects concerning preparation, certification, and issuance of this SRM were coordinated through the Office of Standard Reference Materials by R. W. Seward.

Washington, D.C. 20234
May 9, 1980

George A. Uriano, Chief
Office of Standard Reference Materials

Potassium Iodide Material Testing:

Material homogeneity was tested by measuring the absorbance of 1% aqueous solutions of the KI sampled from the top, middle, and bottom of each bottle. These measurements, at 265, 267, and 270 nm, showed no evidence of inhomogeneity. Tests for moisture content (Karl Fischer method) gave an average of 0.007%. This value was sufficiently small that all certified data are based on weighed samples of the undried, as received, material.

Stability of the material to UV and visible light was tested by exposing the KI salt in a low-actinic glass bottle to radiation from a 3 kW xenon-arc lamp at a distance of 25 cm for four days at room temperature. The average absorbances at five wavelengths between 240 and 270 nm of aqueous solutions of the exposed material were 1.1% lower than those of solutions of the unexposed material. In view of the severity of this test, this change was considered minor. Nonetheless, the 1.1% was included in the overall uncertainty of the certified values.

Potassium Iodide Certification Procedure:

The certified specific absorbances were measured in the NBS Center for Analytical Chemistry high-accuracy spectrophotometer [1,2], equipped with a deuterium lamp and UV averaging sphere [3,4]. Thirteen aqueous solutions of KI with concentrations ranging from 0.03 to 30 g L^{-1} were prepared and measured against distilled water, using standard fused-silica cuvettes with pathlengths between 10.001 and 10.009 mm[b]. The absorbance measurements were performed at the temperature, $t = 23.5 \pm 0.5\,°C$. A 0.2 nm bandpass was used, and wavelength settings were made with an accuracy of 0.05 nm. The measurements were made on at least three concentrations at each wavelength. These concentrations were chosen so that most absorbance readings fell between 0.1 and 1.0 to minimize stray light and bandwidth errors. The data were found to obey Beer's law.

Thus, the equation,

$$\epsilon = A/c\ell \qquad (1)$$

was used to calculate the specific absorbances, ϵ, from the measured absorbances, A, and the known values of concentration, c, and pathlength, ℓ .

Limits to random error (two standard deviations) of these measurements (including the photometric imprecision of the spectrophotometer, errors in the repositioning of cuvettes, concentration errors, pathlength uncertainties, temperature errors, Beer's law uncertainties, and bandwidth errors) were determined to be ± 2.4%. The error due to the 0.05 nm uncertainty of the wavelength calibration of the spectrophotometer is ± 1.1%.

The uncertainty of 5% stated on the face of this certificate represents the sum of these errors and the above mentioned 1.1% uncertainty due to instability of the KI. No corrections were applied for the effects of internal reflections inside the sample and reference cuvettes, nor were the weights corrected to vacuum. These and all other sources of error were considered negligible.

The temperature coefficient at 260 nm was determined to be

$$\frac{1}{\epsilon} \cdot \frac{d\epsilon}{dt} = 0.031\ °C^{-1}. \qquad (2)$$

For precision measurements, it is recommended that SRM 2033 be used in thermostated cuvettes at 23.5 °C, or that a temperature correction according to Eq. (2) be applied.

[b] Issued by NBS as SRM 932, Quartz Cuvette for Spectrophotometry.

Instructions for Use of Potassium Iodide

Storage and Preparation:

SRM 2033 should be stored in the original, low-actinic glass bottle and the cardboard container in which it was issued to protect it from unnecessary exposure to light and humidity. When so stored, the expected stability of this material is at least three years. Thus, until additional stability data are obtained, this material should be used within three years of the date of purchase.

All solutions prepared from SRM 2033 should be made in borosilicate glass containers using distilled water and transfer pipettes (Pasteur type) of the same glass and fitted with rubber bulbs[c]. Use clean spectrophotometer cuvettes, free of scratches, made of non-fluorescent fused silica, and fitted with ground-glass or Teflon stoppers to minimize evaporation; or preferably use NBS SRM 932. Mark all cuvettes to assure the same orientation in the spectrophotometer, and place them into their respective holders. Using transfer pipettes of the type mentiond, rinse each cuvette several times with distilled water. Prepare a solution of KI in distilled water (e.g., 1%), fill the sample cuvette with the KI solution and the reference cuvette with distilled water. Measure the absorbance, leaving the cuvettes in their holders, empty them (using the pipettes), and repeat the rinsing and filling operations until constant absorbance readings are obtained. Fresh solutions should be made before every test.

Measurements:

A 1% KI solution ($c = 10 \text{ g L}^{-1}$) with a 1 cm pathlength exhibits a sharp cutoff in transmittance near 260 nm; i.e., it transmits more than 90% above 273 nm, but less than 0.01% below 258 nm. Therefore, with the monochromator set for a wavelength below 260 nm, any appreciable amount of light detected is heterochromatic stray light, which consists of wavelengths above the cutoff. The amount of stray light in the spectrophotometer at wavelength λ may be determined from the equations:

$$T'(\lambda) = \frac{T(\lambda) + x(\lambda)}{1 + x(\lambda)} \tag{3}$$

$$x(\lambda) = \frac{T'(\lambda) - T(\lambda)}{1 - T'(\lambda)} \tag{4}$$

where:

$x(\lambda)$ is the stray light ratio,

$T'(\lambda)$ is the apparent transmittance,

$T(\lambda)$ is the true transmittance.

The stray light ratio, $x(\lambda)$, is the proportion of heterochromatic stray radiant energy in the spectrophotometer for the wavelength setting λ. Equation (4) forms the theoretical basis for the determination of the stray light ratio by comparing apparent transmittance to true transmittance. For SRM 2033, the true transmittance, $T(\lambda)$, of KI solutions may be calculated as:

$$T(\lambda) = 10^{-\epsilon(\lambda) c \ell}$$

where $\epsilon(\lambda)$ is the certified specific absorbance given on the face of this certificate.

---

[c] Soft glass containers and pipettes contain residual amounts of UV absorbing material, but can be used after proper cleaning. Several rinses, first with isopropyl alcohol and then with distilled water, are generally adequate.

The apparent transmittances of KI solutions depend not only on the stray light ratio of the spectrophotometer, but also on the wavelength accuracy and the spectral bandwidth, which can significantly affect the results obtained because of the steep slope of the absorbance cutoff of KI. A low-pressure mercury discharge lamp is suggested for verifying the wavelength scale, and a bandwidth as narrow as compatible with adequate signal-to-noise ratios should be used.

Optical Attenuator:

Many instruments do not permit the direct measurement of transmittances below certain limits, e.g., 1% or 0.1%. To measure stray light ratios below these limits, the reference beam of the spectrophotometer must be attenuated to extend the transmittance scale into the low-transmittance region.

As mentioned on the face of this Certificate, the optical attenuator consists of two filters, each of which has a nominal transmittance of 10 percent. One filter is mounted in a metal holder that can be inserted in the sample compartment of the spectrophotometer (5). This holder is provided with a front and rear shutter, one of which has a window in which the second filter is mounted. The holder and shutters are flat black. This unit was produced in the NBS Instrument Shops.

Instructions for Use of the Attenuator

This unit can be used to attenuate the incident radiation in the reference beam of the spectrophotometer in two steps by a total factor of about 100, corresponding to about 1 percent transmittance (6). To attenuate by a factor of 10, the attenuator is inserted in the sample compartment of the spectrophotometer facing the incident beam, with both shutters removed. A further attenuation by a factor of about 10 is obtained when the shutter carrying the second filter is inserted in the holder, in front of the first filter. Under these conditions, and with an instrument that cannot measure transmittances lower than 1 percent, the use of the attenuator will permit heterochromatic stray light to be measured down to about 0.01 percent. This attenuation procedure can be used with spectrophotometers capable of scale expansion.

Isochromatic Stray Light:

The radiation attenuator can be used to assess the isochromatic stray light that results from reflection of the incident radiation at the surface of the sample and various optical components, and reaches the photodetector without passing through the sample. It is implicitly assumed that the sample compartment of the spectrophotometer is light-tight.

The measurement is performed by placing the attenuator in the sample beam of the spectrophotometer, with the opaque shutter placed at the rear of the filter holder. Under these conditions, if a signal is detected, it is caused by reflection at the surface of the filter exposed to the incident radiation. This radiation is scattered from the walls and other components of the sample compartment and reaches the photodetector without passing through the sample (5). This signal is the isochromatic stray light.

Isochromatic stray light, which passes through the sample, is generally caused by interreflections between lenses and/or other sample compartment elements. Tests for this stray light component, which is not detected by the methods described above, are discussed in references 7 and 8.

References:

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SRM 2033