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Spectrophotometric Standards

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To be useful, spectrophotometric measurements must be believable and practical. The basic standard for any believable spectrophotometric measurements is the ability to accurately compare fluxes of radiation within the framework of a well-defined geometry. The emphasis in the program proposed for the Institute for Basic Standards is to develop such ability over the broadest range of spectrophotometric measurements. Establishing such a basis will enable the National Bureau of Standards to render real assistance to those who deal with the problem of making practical measurements

Key words: Photometric accuracy; standards, spectrophotometric.

Spectrophotometric standardization is probably best described as elusive. This is not to say that it is an animated being which goes skulking from one hiding place to another, nor is it something which cannot be obtained. Rather, its elusiveness lies in the extreme care required to define what is to be measured and to make sure that something else is not being measured instead. Very seldom is there a one-to-one relationship between the spectrophotometer output and the desired quantity. To standardize a useful spectrophotometric measurement is to determine what *actual* relationship exists.

This assertion can best be illustrated in terms of very high accuracy measurements. The careful worker in spectrophotometry, as it is presently applied, can in general feel confident of an accuracy of plus or minus 0.005 in transmittance in the visible range and, under very favorable circumstances, attain a confidence to 0.002. Very high accuracy measurement for the purposes of this discussion would correspond to confidence to within less than 0.001 in transmittance in the visible, with corresponding degradations in the more difficult wavelength regions and for more difficult geometries. At this level, the effects to be discussed are very important and can determine success or failure in achieving the desired accuracy. It should also be borne in mind that where the accuracy really counts is in determining the desired property of the material being investigated, which is seldom completely or even partially an optical property. If the coupling between the property of interest and the measured spectrophotometric quantity is weak, as is the case in many health and environmental measurements, it may be necessary to achieve a very high spectrophotometric accuracy in order to measure the desired property even crudely. Consider as an example some of the factors which enter into the problem of determining the concentration of a solute in a solution, such as:

a. Linearity—Although it is important to know the linearity of an instrument, knowing it is not sufficient to assure accuracy in measuring transmittance, since many other factors contribute to producing the spectrophotometer output. It should also be noted that gages, such as filters, which have been calibrated on one instrument cannot provide a highly accurate means of determining the linearity of another instrument, since gages have a number of properties which are measured in different ways by different instruments.

b. Image Shift—Since inserting a medium in the beam changes the optical path length, the focus, and hence the way the receiver is illuminated, will be changed. Also, if the sample is tipped, as is sometimes done to reduce the effect of interreflections, the light reaching the receiver will be shifted laterally. If the receiver is at all sensitive to such variations in illumination, the spectrophotometer will measure this effect as well as transmittance.

c. Reflections—In the usual arrangement of a solution in a cuvette, there will be four surfaces at which reflections occur, and multiple reflections between these surfaces contribute to the spectrophotometer output in most cases. If there are any transmitting glasses such as lenses or windows in the spectrophotometer, there can be additional effects due to reflections from these.

d. Index of Refraction—The index of refraction of a solution fluctuates strongly in the wavelength range occupied by an absorption line. This change in refractive index will in turn cause image shifts and changes in reflections within that range of wavelengths.

This again will appear as a part of the spectrophotometer output.

e. Identification of properties—Even if a spectrophotometer were made to measure transmittance perfectly, transmittance per se is often not the property of interest. Transmittance is a complex quantity which depends upon the measurement geometry and several properties of the sample, and thus is not simply related to a single property such as the concentration of a solute in a solution.

The foregoing discussion has dealt with some of the interactions between spectrophotometer and test sample. There are, of course, other sources of difficulty such as scattering of light by the sample, changes in the sample with temperature and stray light from the monochromator. All of these also contribute to the spectrophotometer output.

The point to be made here is that if high spectrophotometric accuracy is to be achieved, these effects must be accounted for. The electronics, optics, detectors and readouts of the spectrophotometers available have been greatly improved, so that these instruments can be made much more stable and more sensitive than they were five or ten years ago. The result has been more than an order of magnitude improvement in precision, but there has been no corresponding improvement in accuracy. Improving accuracy can only come about through concerted efforts by instrument makers, standards laboratories, and those who need to perform very accurate spectrophotometric measurements.

Instrument makers should at the very least make the optical layout of each instrument known *in complete detail* so that the owner or prospective owner can see what stands in the way of his achieving the accuracy he needs. It would also be a great contribution if each new instrument intended for high accuracy work were designed for accuracy in a particular type of measurement, without undue concern that it give the same "numbers" for a given filter that last year's instrument gave.

The standards laboratory has a many-faceted role in standardization for high accuracy. Research should be conducted to find how the magnitude of each of the various contributions to error can be determined, to find in which case corrections can be made, and in such cases give practical procedures for making the corrections. When special gages are needed to aid in a particular step of calibration, the standards laboratories should see that these are available and that all the pertinent properties of these gages are made known. For example, for very high accuracy work, the absorbance, index of refraction, thickness and scattering properties of a filter should be supplied by the standards laboratory instead of just a transmittance value. For such work it makes no sense for a standards laboratory to pass out gages with a "best value" of some single property attached. The standards laboratory must maintain a versatile, thoroughly evaluated, and carefully documented measurement system in order to perform its own work and evaluate the work of others. Also, the standards laboratory must constantly be aware of new developments in spectrophotometry and maintain a close liaison with all groups involved in the measurements in order to assist everyone, including the standards laboratory itself, to achieve the needed measurements.

The ultimate responsibility for standardization of very high accuracy measurements lies in the person or persons performing the measurements. The user must take the steps necessary to determine what stands in the way of his obtaining the particular data he desires with his particular instrument and make appropriate corrections. The operator of the spectrophotometer may have the knowledge and experience to do this on his own, or he may have to rely heavily on instructions provided by others, but he is the one who must standardize his measurements.

What has been described is spectrophotometric standardization for very high accuracy—standardization for the future, since very high accuracy spectrophotometry is not being used at the present. However, factors such as those listed in the early part of this discussion contribute to any measurement, and they should be considered when making measurements at any level of accuracy. The cost in effort to obtain very high accuracy measurements will be great, so it is extremely important to attempt them only where the anticipated benefits will be correspondingly great—they certainly should not be attempted when not really needed. Very high accuracy measurements *can* be obtained with diligence on the part of those making the measurements, and support for such measurements *can* be provided by instrument makers and standards laboratories, but a change in attitude and approach by all may be required before they are achieved.

To be useful, spectrophotometric measurements must be believable and practical. The basic standard for any believable spectrophotometric measurements is the ability to accurately compare fluxes of radiation within the framework of a well-defined geometry. The ability to relate the results of such measurements to a property which is to be determined can only be achieved through careful interpretation of the spectrophotometric output based on considerations such as those described above. The emphasis in the program being proposed for the Institute for Basic Standards is to develop a strong measurement capability over the broadest possible range of spectrophotometric measurements. Establishing such a capability will enable the National Bureau of Standards to render real assistance to those who deal with the problem of making practical measurements for a given application.

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