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Development and Certification of NIST Standard Reference Materials for Relative Raman Intensity Calibration

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Abstract. In analytical Raman spectroscopy it becomes increasingly important to employ a procedure for the correction of the relative intensity of Raman spectra. The determination of the intensity response function of a Raman instrument traditionally has been carried out through a white light source that has been calibrated for its relative spectral irradiance. While this method will furnish a correction curve to yield spectra corrected to relative Raman intensity, it is often cumbersome and fraught with experimental difficulties that can profoundly affect the reliability of the correction procedure. An alternate methodology that permits a simplified calibration of the Raman instrument response function is based on the use of luminescent glass standards that transfer a white light calibration onto the Raman measurement system. In this procedure, a measurement of the luminescence of an intensity standard, whose relative irradiance has been determined, provides a means to establish the instrument response function. Correction of measured spectra by this function furnishes spectra that are free of instrumental intensity artifacts. Based on this approach, NIST is developing a series of Standard Reference Materials (SRMs) for the calibration of Raman intensity. This process, and the results obtained thereby, is described for Raman spectroscopy measurements employing 785 nm excitation. The procedure is valid for both macro-sampling and micro-sampling Raman work.

Key words: Raman spectroscopy; intensity calibration; intensity standards; instrument response function; luminescent glasses; Standard Reference Materials (SRMs).

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Introduction and Background

Attention has been given in recent years to the various issues that arise from the general lack of standardized procedures for the calibration of Raman spectral intensity [1–7]. With the increasing acceptance of analytical Raman methods and the availability of a variety of commercial Raman systems, it becomes important that uniform calibration methods be developed that provide some means for the correction of the Raman instrument response function. As matters stand now, Raman spectra are typically not corrected for instrument-dependent variations in spectrometer sensitivity across any chosen Raman shift range. Especially for measurements at different excitation wavelengths, this leads to an often severe distortion of relative peak intensities which impedes calibration transfer and affects spectral matching in library search routines. These issues have been brought forth by the McCreery Group at The Ohio State University [4–6]

to the point where national standards organizations (e.g., ASTM, NIST), federal regulatory agencies (such as the Food and Drug Administration), and the analytical Raman community (especially the instrument manufacturers) have taken note.

We are reporting here on the development, at the U.S. National Institute of Standards and Technology (NIST), of a series of secondary standards for the calibration of the relative Raman spectral intensity. The calibration procedure is based on the use of luminescent glasses, doped with either transition metal or rare earth ions, that furnish broad-band emission spectra upon laser excitation, over any given Raman shift range [7–9]. In principle, each standard selected for a specific Raman excitation wavelength will have a unique luminescence emission profile that depends both on the composition of the glass and the laser wavelength. The intensity of the luminescence spectrum is calibrated versus the radiant output of a NIST-calibrated white light source, thereby transferring the white light calibration onto the output emission of the luminescent glass standard. Since the glass standard is measured in the Raman instrument under the same conditions as is the analytical sample, the spectrum of the standard is subject to the same instrumental response function. The known (i.e., intensity corrected) spectrum of the luminescent standard is expressed as a polynomial which is used in a straightforward computational procedure to yield an intensity correction curve. This correction curve is unique to each Raman system and is used to correct the measured Raman spectrum of the sample to provide a spectrum that is free of instrumental intensity artifacts. Correction for instrumental response is particularly important in Raman work using near-infrared (NIR) lasers and charge-coupled device (CCD) detectors due to the widely varying response function, depending on the type and quantum efficiency of the CCD used [5, 6]. This leads to very different intensity profiles for sample spectra excited at 785 nm vs. 514.5 nm, as illustrated in Ref. [5].

The importance of the Raman intensity correction can be demonstrated with the example of the Raman spectrum shown in Fig. 1 of benzonitrile (C_6H_5CN), a simple organic liquid, excited at 785 nm. The spectrum is well known, and all of the bands have been assigned to fundamental vibrations of the molecule. In the corrected spectrum, the most intense bands are at $\sim$998 cm^{-1} (benzene ring ''breathing''), at $\sim$1596 cm^{-1} (ring stretches), at $\sim$2228 cm^{-1} (–CN, nitrile stretch), and at $\sim$3069 cm^{-1}. (aromatic all C–H stretch). Of these, the nitrile band is the second most intense feature, and the all-CH band is of moderate intensity. In the measured uncorrected spectrum, these same relative intensities do not exist. The corresponding bands are at the same Raman wavenumbers but the band intensities are grossly distorted, especially for the bands beyond 1200 cm^{-1} (Raman shift). This is most exacerbated for the all-CH band, which is just a blip in the uncorrected spectrum, obviously just barely detected. Consequently, the relative band intensity ratios are not the same for the corrected spectrum vs. the uncorrected, measured spectrum. This, of course, is the result of the instrument response fall-off in the 785 nm Raman measurement due in large part to the much-diminished detector sensitivity at Raman shifts beyond $\sim$2500 cm^{-1}.

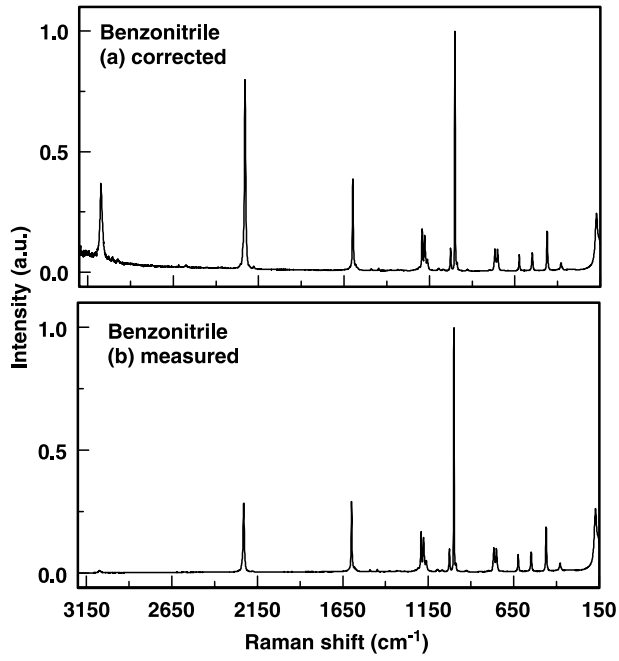

Fig. 1. Raman spectrum of (liquid) benzonitrile excited at 785 nm. The top spectrum (a) is the spectrum corrected to ''true'' relative intensity. The bottom spectrum (b) is the trace as recorded by the macro-Raman system, prior to intensity correction

The advantages of the use of luminescent standards for Raman intensity calibration were recognized also by the Raman Subcommittee of the American Society for Testing and Materials (ASTM, Subcommittee E13.08 on Raman Spectroscopy), charged to look into the problems of Raman intensity calibration. This standards body, in turn, recommended that NIST respond to the needs of Raman spectroscopists, the instrument manufacturers, and those of a cross-section

of the regulated U.S. industries, by undertaking a research program aimed at developing a series of Standard Reference Materials (SRMs) for the calibration of the Raman intensity [9]. NIST has a long history of providing standards for both physical and chemical measurements [10, 11], and this projected series of Raman SRMs will in fact be the first to address the needs of the analytical Raman community [12].

In this paper we describe the development and the procedures used for the measurement and certification of a series of luminescent glasses intended to serve as secondary standards for the calibration of the relative Raman intensity. This work has resulted in the issuance by NIST, in June 2002, of SRM 2241 as the standard for Raman spectroscopy using 785 nm laser excitation. Work is in progress on additional SRMs for this purpose, and these will cover the Raman spectral regions defined by laser excitation at 488.0, 514.5, 532.0, 633.0 and 1064.0 nm, the latter wavelength generally employed by commercial Fourier-Transform (FT) Raman systems.

Luminescent Glasses for Instrument Response Function Correction

Leading up to this work on the development of Raman intensity standards, we have examined the fluorescence emissions, excited at common Raman laser wavelengths, of various fluorescent ions present in several types of glass matrices [7]. Of specific interest are glasses bearing one or more rare-earth (RE) elements and/or certain transition metal or actinide ions. The laser-excited photoluminescence emissions from glasses doped with such fluorophores can be tailored through the composition and structure of the glass. As a result, glasses of this type, by virtue of their luminescent emissions, can serve as ideal secondary radiometric sources for the calibration of the instrument response function. Many of the RE ions (e.g., Er^{3+}, Eu^{3+}, Nd^{3+}), doped into a glass matrix, produce interesting fluorescence spectra that show strong resemblance to the spectrum of the corresponding oxide (e.g., Er_2O_3, Eu_2O_3, Nd_2O_3), typically existing as the crystalline solid. While the spectra of these ionic solids can often be quite complex, consisting of several, well-resolved bands (as measured in a Raman instrument), the spectra of these RE ions present in a glass matrix are usually less complex, due to the merging of narrow bands to result in broadened spectral bands [13, 14]. Typically, the spectral emissions of RE ions in glass matrices are sensitively affected by the type of the glass (e.g., silicate, borate, phosphate) and the state of order or disorder of the glass matrix. The luminescence spectra of RE-bearing glasses, depending on RE concentration and quenching effects, are often intense and may consist of both narrow-band emissions and/or broad-band emissions covering a wide spectral range. We have examined the spectra of such glasses across the visible (VIS) region through the near-infrared (NIR) region, with Raman laser wavelengths from 488.0 nm (Ar-ion laser) to 1064 nm (Nd:YAG laser, FT-Raman system). In this work, we have identified stable glass compositions with either incorporated RE ions or heavy metal ions that produce featureless broad-band fluorescence emissions upon laser excitation. We have studied these emissions for a series of RE ions and a number of metal ions (e.g., Fe^{3+}, Ni^{3+}, Cr^{3+}, Cu^{2+}, Mn^{4+}, UO_2^{2+}), present at low concentrations in several glass matrices, principally silicate, borate, phosphate, and germanate. Importantly, these glasses have been investigated in both macro- and micro-sampling measurements and are found to be homogeneous at the micron-scale. One of the goals of the NIST work has been to identify doped glasses that would exhibit great resistance to photobleaching upon prolonged laser irradiation.

Luminescent Glass Standard for Raman Work at 785 nm Excitation

Early on it was realized that the intensity correction was most critical for analytical Raman work utilizing 785 nm excitation (c.f. Fig. 1). The decision to start out with a standard for this 785 nm Raman regime was supported by the ASTM Raman Subcommittee, based on input from the larger Raman community. Lasers operating at 785 nm are widely used in industry, and the spectra obtained from such systems are the most in need of relative intensity correction. In the Raman region defined by 785 nm excitation, the typical CCD detectors are characterized by a widely varying response function, depending on the quantum efficiency ($Q(\lambda)$) of the CCD used. Typically, there is an increase in $Q(\lambda)$ for all such detectors over the usual Raman spectral range of 100–3600 cm^{-1} (Raman shift) relative to 514.5 nm, whereas there is a sharp decrease in $Q(\lambda)$ over that same Raman range for 785 nm excitation where CCD detectors nearly lose

all sensitivity at the far end of this range. The correction curve, in the latter case, will show a sharp upward trend toward the longer wavelengths of the Raman range to account for this diminished sensitivity response, unless other instrumental effects partially offset this trend.

The correction procedures established at NIST for work with 785 nm excitation have focused on the use of a series of Cr-doped glasses that possess the properties required, namely that they be stable, homogeneous, immune to photobleaching by the laser, and furnish a suitable broad-band luminescence spectrum. These criteria are well met by a number of glass compositions that bear Cr^{3+} at concentrations in the range of 0.1–0.005 mole% Cr_2O_3 in a variety of suitable, robust and stable glass matrices [15, 16]. One of the exploratory glasses examined for its luminescence properties was Glass K-3653, an in-house NIST research glass, with very low chromium content (0.005 mol% Cr_2O_3), doped into a Na–Ba–Zn-silicate matrix known to be very stable. This glass, when excited at 785 nm, under quite moderate laser power densities, emits a non-bleaching, broad- band luminescence spectrum, shown in Fig. 2. The intensity correction to the measured spectrum was made through the calibrated spectral output of a white light irradiance source (spectral lamp). Note the differing shape of the two curves and the fact that the luminescence peak of the white-light (WL) corrected spectrum now is observed at higher Raman shift. Clearly, the intensity correction leads to an emission profile quite different from that seen in the measured, uncorrected spectrum.

In the selection of a suitable luminescent glass for intensity correction at 785 nm, it is especially useful if the glass spectrum maintains good relative intensity in the region of Raman shift beyond 2000 cm^{-1}. The corrected luminescence spectrum of Glass K-3653 has its peak maximum at ~1700 cm^{-1}, beyond which its intensity gradually drops off. Cr-doped glasses of other compositions show luminescence curves for which the band maximum is shifted to longer wavelengths, i.e. to higher Raman shifts. This is more desirable for the calibration procedure as it optimizes the fidelity of the intensity correction. For this reason, it was decided to adopt a chromium glass composition based on a simple, ternary sodium borosilicate matrix: SiO_2 (67.35 wt%), Na_2O (6.95 wt%), B_2O_3 (25.65 wt%). The chromium concentration of this glass is 0.05 wt% (or 0.02 mol%) Cr_2O_3. This glass is the certified Raman intensity standard, designated SRM 2241. The peak of its luminescence spectrum (when intensity corrected) now falls at approximately 2600 cm^{-1}, and thus is considerably shifted to higher Raman frequencies.

Determination of the Relative Spectral Irradiance of SRM 2241

The relative spectral irradiance of the glass standard (SRM 2241) was determined through a calibration of the instrument response against a secondary white

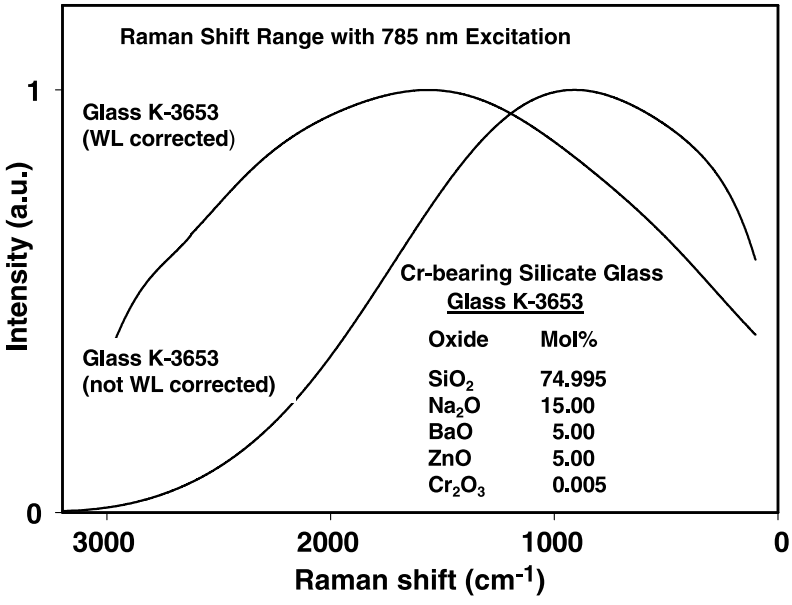

Fig. 2. The broad-band luminescence spectrum of Cr-bearing silicate Glass K-3653, excited at 785 nm. The spectrum is plotted as the intensity corrected spectrum (white light corrected) and the measured spectrum (not white light corrected). Both curves are normalized to unity in the peak of the band

light source of known relative irradiance. These procedures were carried out on three different Raman systems and utilized two interchangeable secondary white light emission sources (each an integrating sphere source fitted with a tungsten-halogen lamp) that had been calibrated for its relative irradiance by NIST's Optical Technology Division Calibration Services program.

The three Raman spectrometers used in this work provided us with a variety of measurement conditions thought to be typical of those encountered by analysts in the Raman spectroscopy field. One of the Raman systems was a commercial Raman microscope employing a deep-depletion, front-illuminated, red-enhanced CCD detector. The other two were macro-sampling Raman systems employing commercially-available, single-grating, 0.5 m focal-length imaging spectrographs of Czerny-Turner design. These spectrographs used back-illuminated CCD's.

In the macro-Raman fore-optics, a separate laser focusing lens and a separate scattering collection lens were used, each independently adjustable. All three Raman systems utilized the same Ar-ion pumped titanium:sapphire laser optically tuned to emit 785 nm radiation. The holographic notch filter used with each of these Raman systems allowed the spectrum to be collected within 150 cm^{-1} of the laser excitation line. A depolarizer was used in the collimated portion of the exit beam, ahead of the spectrometer entrance slit, to scramble any polarization either from the sample or induced by the collection optics. In all cases, the spectra were collected in a back-scattering configuration and were recorded with a spectral resolution of 3 cm^{-1}. Raman shift calibration was performed using the literature values of the emission lines of neon.

Measurements were made on the three instruments described and an intensity-correction applied to the measured luminescence spectra. A statistical analysis was performed on a set of twenty-three spectra, derived from over 100 measurements. Figure 3 shows certified fifth-order polynomial that describes the intensity-corrected luminescence spectrum of SRM 2241, plotted over the Raman range 250–3350 cm^{-1}. This polynomial is certified for use over the temperature range 20 °C to 25 °C. Shown with this curve are the associated 95% confidence curves (CC). The certified coefficients of the polynomial and their respective uncertainties are listed in the NIST Certificate of Analysis [17].

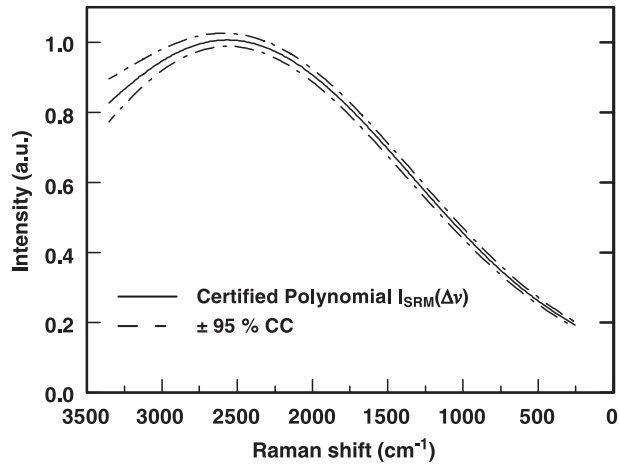

Fig. 3. Spectral curve defined by the fifth-order Certified Polynomial and its 2σ error bounds (i.e., the 95% confidence curves) describing the luminescence spectrum of SRM 2241 over the temperature range of 20 °C to 25 °C. The peak of the curve is normalized to unity. The x-axis is in Raman shift relative to 785 nm excitation

In the use of the SRM glass by the analyst, the luminescence spectrum of the standard is measured using the same instrument settings and parameters as are used for the analytical sample. The intensity correction curve for a given Raman system is generated numerically by dividing the polynomial curve of SRM 2241 by the measured luminescence spectrum of the SRM glass, for each data point, across the certified spectral range:

$$\text{Intensity Correction Curve} = \frac{\text{Certified Spectrum of SRM 2241}}{\text{Measured Spectrum of SRM 2241}}$$

Multiplication of the Raman spectrum of the analytical sample by this curve, point by point, then corrects for the instrumental intensity artifacts present in the measured spectrum. The intensity correction curve, derived in this procedure for the micro-Raman system used in these certification measurements, is shown in Fig. 4. It is specific to this system, the Raman microscope, as it compensates for the relative intensity distortions due to the focusing/collection objective, all other fore-optical components, the specific grating used in the spectrometer, and the specific CCD detector employed. Merely the change to another focusing objective (e.g., from a 10 × objective to a 50 × objective) may slightly alter the shape of the correction curve. More profound changes can be expected as spectrometer gratings are switched from one measurement to another, with all other conditions remaining unchanged.

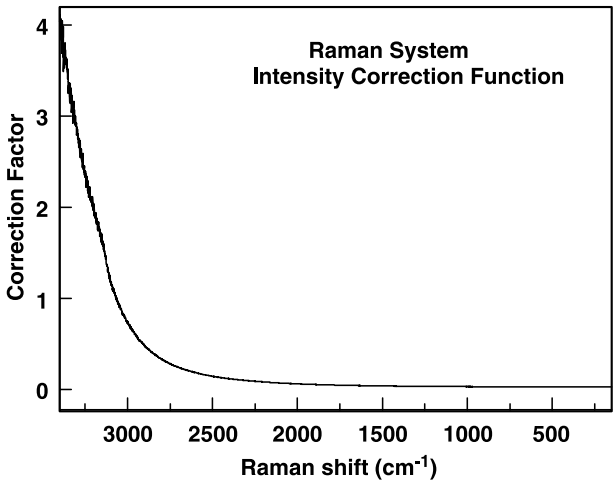

Fig. 4. Relative intensity correction curve for the micro-Raman system (a commercial Raman microscope) used in the certification measurements of SRM 2241, employing Raman excitation at 785 nm. Values of the correction factor are scaled to the indicated values. The shape of the curve is unique to the Raman system

The SRM is intended for use in measurements over the range of 20 °C to 25 °C with Raman systems employing (785 ± 1) nm laser excitation. The photostability of SRM-2241 was assessed through tests exposing the glass to high-intensity (irradiance of approx. 1 MW cm^{-2}), focused 785 nm laser light, continuously for a period of about 1.5 hrs. No detectable bleaching of the glass luminescence signal was observed under these conditions. The SRM Certificate gives detail on the certified values of the glass polynomial and their uncertainty [17]. It also provides reference values of the polynomial for use of the SRM over the broader temperature range of 15 °C to 30 °C. Further given are instructions for use of the standard and an equation that expresses the shape of the luminescence spectrum of the SRM on the wavelength scale. This coordinate transformation allows for the intensity correction of luminescence or fluorescence spectra where the x-axis is expressed in wavelength (nm) units.

Validation of Raman Intensity Correction through SRM 2241

It is useful to have a procedure that provides a validation of the intensity correction curve that has been determined for a given Raman instrument and that will furnish information on the reproducibility of the intensity correction procedure. This can be accomplished by determining the peak area ratios of the intensity corrected Raman spectrum of cyclohexane [6]. Cyclohexane has been designated by the ASTM as a standard for the calibration of the Raman wavenumber, together with a suite of other readily available organic compounds [18]. This liquid has a simple Raman spectrum with five well-defined bands that lend themselves to unambiguous and straightforward peak area integration. Integrated peak areas are typically determined by standard spectroscopic software that permits the user to assign the integration limits and to employ an appropriate method for treating the spectral baseline. Generally, the band areas are normalized to that of a prominent peak in the spectrum, and the band area ratios are calculated as a quantitative measure of the relative band intensities of a given sample spectrum. This procedure is readily carried out for cyclohexane, the spectrum of which has been used as a de facto standard for the computation of Raman band intensity ratios [5, 6]. For cyclohexane, the five Raman peaks (and the range of integration) considered in this procedure are the bands centered at 802 cm^{-1} (700 to 900 cm^{-1}), 1028 cm^{-1} (925 to 1125 cm^{-1}), 1267 cm^{-1} (1180 to 1315 cm^{-1}), 1444 cm^{-1} (1380 to 1525 cm^{-1}), and all-CH (2587 to 3068 cm^{-1}). Figure 5 shows the measured and intensity corrected spectrum of cyclohexane, excited

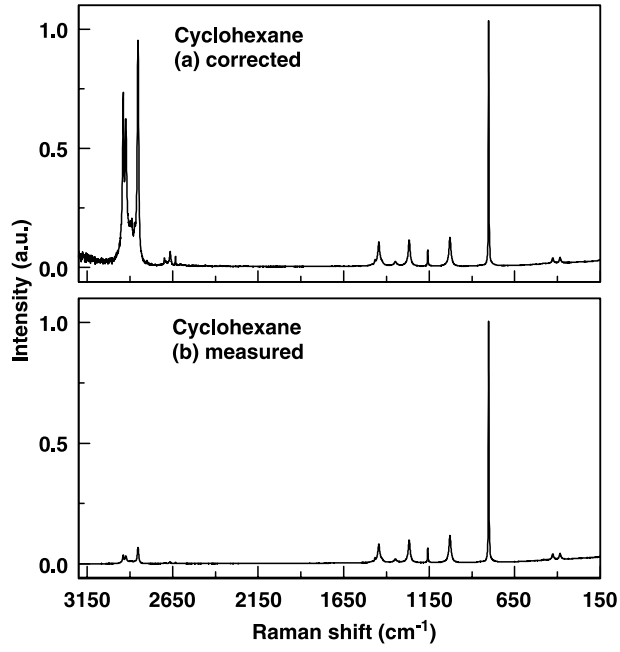

Fig. 5. Measured and intensity-corrected spectrum of cyclohexane, excited at 785 nm. The corrected spectrum was obtained through the use of intensity standard SRM 2241 and the intensity correction curve derived thereby. Both spectra are normalized to unity in the peak of the 802 cm^{-1} band

at 785 nm, obtained with the micro-Raman system. Typically, the band areas are ratioed to the area of the sharp 802 cm^{-1} peak. For 785 nm excitation, the values of these band area ratios show a range of variabilities, and it is the all-CH band ratio which usually shows the greater variability due to the intrinsic uncertainties associated with the measurement of the Raman signal in the high Raman shift range [6, 12].

NIST, as part of its program to develop relative intensity standards for Raman spectroscopy, carried out a round-robin of measurements with members of the ASTM Subcommittee E13.08 on Raman spectroscopy. The object was to learn how well the proposed correction procedure would be handled by analysts in the field. This round-robin circulated pieces of a luminescent Cr-glass, a prototype of SRM 2241, for which the known relative irradiance (under 785 nm laser excitation) had been determined by NIST, and was described by a polynomial that was made available to the user. As part of this exercise, the Raman spectrum of cyclohexane was measured. The relative intensity of that spectrum was corrected through the measurement of the glass luminescence, and the peak area ratios of cyclohexane were calculated. The results of this round-robin were reported in Ref. [19].

Since then, NIST has done further studies on the systematic effects on measurements of Raman spectra. Measurements of the cyclohexane spectrum, using 785 nm excitation, were performed with both a macro-Raman system, with a depth-of-focus of about 1 mm, and with a Raman microprobe system which, using 10× and 20× microscope objectives, had considerably shorter depth-of-focus. These measurements were done using a standard 10 mm quartz cuvette. For the macro-system, large systematic changes in the peak area ratio of the CH-stretch region to the 802 cm^{-1} peak could be observed as the collection lens focal point was moved from the front window of the cuvette to the back window of the cuvette. Table 1 indicates the total variability of the peak area ratio measurements of cyclohexane that we have observed from two dozen independent measurements done over a period of three months. The data were obtained with a macro-Raman system, based on measurements employing a 10 mm cuvette and excitation at 785 nm.

Table 1. Intensity corrected spectrum of cyclohexane measured in a 10 mm quartz cuvette (excitation at 785 nm)

Computed band area ratios ($R = I_x/I_{802}$)		
Band position (cm^{-1})	Integration range (cm^{-1})	Range of band area ratios ($R = I_x/I_{802}$)
802	700 to 900	1.00
1028	925 to 1125	0.54 to 0.60
1267	1180 to 1315	0.41 to 0.49
1444	1380 to 1525	0.52 to 0.61
2900	2587 to 3068	6.0 to 10.0

The differences in the band area ratios shown in Table 1 are considerably larger than the reproducibility of repeated measurements at a given depth of focus into the cuvette. Similar effects could be observed with the micro-Raman system, but the observed changes were not as large, at least in part because the limited depth-of-focus did not allow collection of the Raman signal from deep into the cuvette. Effects of diffraction and refraction on the collected Raman signal have been the focus of recent studies [20, 21] and may account for some of these systematic variabilities in the measured band area ratios. However, it should be noted that for purposes of checking instrument performance, for repeated measurements that are done in a carefully controlled manner, the precision of the measurements should provide a test for the reproducibility of the relative intensity calibration procedure. These NIST studies will be published in more detail when they are complete.

Intensity Corrected Spectra in Micro-Raman Measurements

The application to micro-Raman measurements, of the Raman intensity correction procedure described here, is straightforward. Generally, standard microscope objectives are used, with high collection efficiencies, providing a range of magnifications. This results in a range of focal lengths so that for the higher magnification objectives (20× to 50×) only the near-surface layers of the sample are probed. We should note that the upper surface of the SRM glass slide is frosted, i.e., roughened to a uniform finish, so that the scattering (i.e., the signal) is coming from the roughened surface of the glass. This has the effect of diminishing the depth penetration of the focused laser and collecting the glass luminescence spectrum essentially from the top surface of the glass slide.

The following two examples demonstrate the implementation of the Raman intensity correction procedure, with SRM 2241, on our commercial Raman microscope using 785 nm excitation. The first is the result of a measurement on a sample of thin-film polyethylene. This spectrum is shown in Fig. 6. The second

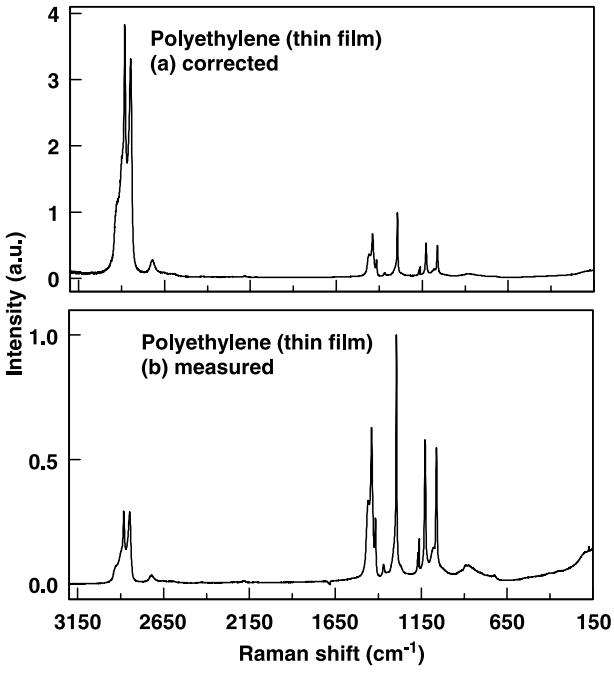

Fig. 6. Micro-Raman spectrum of a thin film (thickness: 45 μm) of low-density polyethylene, excited at 785 nm. Shown are the measured spectrum and the intensity-corrected spectrum, with the correction made through SRM 2241

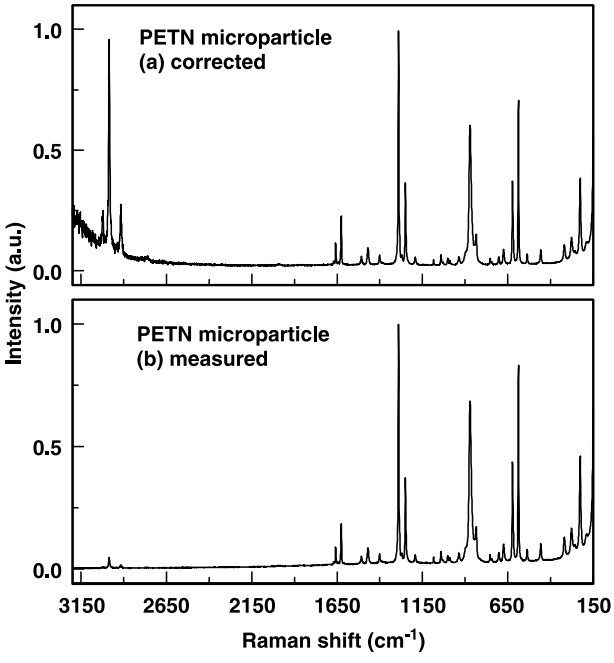

Fig. 7. Micro-Raman spectrum of a microcrystallite (size: 20 μm) of the explosive PETN (pentaerythritol tetranitrate) excited at 785 nm. Shown are the measured spectrum and the intensity corrected spectrum obtained through the use of SRM 2241

is the spectrum, shown in Fig. 7, of a crystalline microparticle of the explosive PETN (pentaerythritol tetranitrate). Both microsamples were analyzed under standard, routine conditions in the Raman microscope, and the luminescence spectrum of the SRM 2241 glass slide was acquired under the same conditions. In the uncorrected spectrum of the polymer film, the sharp band at 1295 cm^{-1} is the dominant (in intensity) band; however, the aliphatic C–H stretching band, at ~2870 cm^{-1}, is of much diminished intensity, in this spectral regime of low signal sensitivity. Correction of the measured intensity of the spectrum makes the C–H band the dominant spectral feature, so that this has now become the "conventional" spectrum as it appears in Raman spectral libraries.

The micro-Raman spectrum of the microparticle of PETN shows the corresponding effects that result from the spectral changes associated with the intensity correction procedure. In the uncorrected spectrum, the C–H band (a triplet) is just barely observed. Once the correction is performed, this band is of comparable intensity to the features below ~1700 cm^{-1}. The spectral intensity in other parts of the spectrum, namely the region below ~1700 cm^{-1}, is by far not affected to the same degree. The intensity corrected spectrum of PETN is now in reasonable agreement with the spectrum of this explosive reported in the literature for a wide range of laser excitation wavelengths (e.g., 514.5, 632.8, and 1064 nm) [22–24]. In some cases the authors show the PETN spectrum as an intensity corrected spectrum, as is the case where an instrument response correction has been made for the FT-Raman instrument operated at 1064 nm [22]. In other cases the reported PETN spectra clearly are not intensity corrected, having been obtained under conditions of 514.5 nm and 632.8 nm laser excitation [23, 24]. This is merely indicative of the contemporary state of Raman spectroscopy practice where in the majority of cases spectra are not corrected to their real relative intensities [12].

We can summarize the various merits that define the Raman intensity correction procedure based on the use of SRM-2241 and all other forthcoming luminescent glass standards developed for the same purpose:

1. The glass standards are simple to use, readily adaptable to all types of Raman instrumentation, are relatively inexpensive (current cost, per SRM unit: US $800), and require no calibration over their life time (period of certification: 5 yrs).

2. In use, the Raman standard reproduces the Raman sampling geometry and requires no additional equipment other than what is needed to take the Raman spectrum of the sample.
3. The standards are based on simple glass matrices, doped with a metal ion that is a stable fluorophore with great resistance to laser bleaching. They are of homogeneous composition and luminescent response and have excellent chemical and long-term stability.
4. As luminescent glasses, the standards provide broad, featureless spectral output over the relevant Raman range, readily related to the spectral output of a primary radiometric white light source.
5. Used as secondary standards that provide a source of known relative spectral irradiance, the NIST Raman SRMs allow for the correction of the relative intensity of Raman spectra, thereby furnishing spectra that are instrument-independent, permit the intercomparison of instruments, and provide compatibility with standardized Raman spectral libraries.
6. Finally, the instrument response calibration performed with these Raman standards permits the periodic verification of instrument performance over extended time, an aspect that can be important in quality control regiments.

Conclusions

Described is current NIST work intended to provide a series of Standard Reference Materials which will permit the correction of the measured Raman spectral intensities for the effects of instrumental intensity artifacts. This procedure is based on the use of a set of luminescent glass standards developed for each of the common Raman spectroscopy laser wavelengths. In essence, this procedure entails the transfer of the known relative irradiance of a calibrated white light source onto the luminescent emission of the glass standard. The measurement of the standard by the analyst provides an intensity correction curve, a mathematical function that describes the spectral response behavior of the system, and is specific to a given Raman instrument. Use of the correction curve, in a simple numerical procedure, allows for the correction of the measured Raman spectrum to remove the spectral distortions that result from instrumental intensity artifacts.

The first intensity standard (SRM 2241, for 785 nm excitation) was issued in June 2002 and is available through the Standard Reference Materials Program of NIST [25]. The second standard in this series (SRM 2242, for 532 nm excitation) became available in January 2004. Currently, other standards of this series are under development for Raman work at laser wavelengths 488.0 nm, 514.5 nm, and 1064 nm. The standards are easy to use and permit the routine correction of the relative Raman intensities of spectra obtained from widely varying Raman instrumentation. These merits are expected to stimulate the creation of instrument-independent spectral libraries. Further, this development may bring a more widespread acceptance of Raman spectroscopy as a practical and user-friendly analytical technique.

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References

[1] Purcell F J, Kaminski R, Russavage E (1980) Appl Spectrosc 34: 323
[2] Hamaguchi H (1988) Appl Spectrosc Rev 24: 137
[3] Petty C J, Warnes G M, Hendra P J, Judkins M (1991) Spectrochim Acta Part A 47: 1170
[4] Fryling M, Frank C J, McCreery R L (1993) Appl Spectrosc 47: 1965
[5] Ray K G, McCreery R L (1997) Appl Spectrosc 51: 108
[6] Frost K J, McCreery R L (1998) Appl Spectrosc 52: 1614
[7] Etz E S, Choquette S J, Hurst W S, Blackburn D H (1998) Microscopy and Microanalysis 4 [Suppl] 2: 508
[8] Etz E S, Hurst W S, Choquette S J, Blackburn D H (1999) Microscopy and Microanalysis 5 [Suppl] 2: 58
[9] Etz E S, Hurst W S, Choquette S J (2000) Raman intensity calibration with glass luminescence standards. Microbeam Analysis 2000, Inst Physics Conf Ser No 165. In: Williams D B, Shimizu R (eds) Institute of Physics Publishing, Bristol UK
[10] Rasberry S D (2001) Accred Qual Assur 6: 95
[11] Rasberry S D (2001) Accred Qual Assure 6: 252
[12] McCreery R L (2000) Raman spectroscopy for chemical analysis, chapter 8. John Wiley & Sons, New York
[13] Baesso M L, Bento A C, Miranda L C M, DeSouza D F, Sampaio J A, Nunes L A O (2000) J Non-Cryst Solids 288: 56
[14] Li H, Li L, Vienna J D, Quian M, Wang Z, Darab J G, Peeler D K (2000) J Non-Cryst Solids 278: 35
[15] Strek W, Deren P J, Lukowiak E, Hanuza J, Drulis H, Bednarkiewicz A, Gaishun V (2001) J Non-Cryst Solids 288: 56
[16] Russell D L, Holliday K, Grinberg M, Hollis D B, (1999) Phys Rev B59(21): 13712

[17] Website to NIST SRM 2241 Certificate: https://srmors.nist.gov/certificates/2241.pdf
[18] Annual Book of ASTM Standards – 2002, Volume 03.06, Standard E 1840-96: Standard Guide for Raman Shift Standards for Spectrometer Calibration, ASTM International, West Conshohocken, PA, p 19428
[19] Choquette S, Etz E, Hurst W, Blackburn D (2002) American Pharmaceutical Rev 6(2): 74
[20] Baldwin K J, Batchelder D N (2001) Appl Spectrosc 55: 517
[21] Everall N J (2000) Appl Spectrosc 54: 1515
[22] Akhavan J (1991) Spectrochim Acta Part A 47: 1247
[23] Carter F W S, Sinclair T J (1983) J Raman Spectrosc 14: 410
[24] Cheng C, Kirkbride T E, Batchelder D N, Lacey R J, Sheldon T G (1995) J Forensic Sciences 40: 31
[25] Information on NIST Raman Intensity SRMs, including the SRM Certificates, may be obtained through the NIST website: http://www.nist.gov/srm

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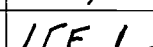
Differentiation of several errors in the same paragraph		There are many faults in this lane	r/ m/ aF i L i J
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