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The Evaluation of Two Commercially Available, Portable Raman Systems

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Abstract: Raman spectroscopy and surface enhanced Raman spectroscopy (SERS) have many attributes that make them attractive for field detection of environmental contaminants, industrial process control, as well as materials detection/identification in agriculture, pharmaceuticals, law enforcement/first responders, geology, and archeology. However, portable, robust, inexpensive Raman systems are required for these applications. In this communication, the performances of two commercially available, portable Raman systems are evaluated.

Keywords: portable Raman systems, SERS, Raman spectroscopy

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Introduction

Raman spectroscopy is an emission technique that involves inelastic scattering of incident laser energy resulting in spectral peaks which are frequency shifted from the incident energy. The Raman spectral peaks arise from changes in polarizability in a molecule during vibration. As a result, virtually all organic and inorganic molecules display a characteristic Raman emission. Because of the high resolution of Raman spectra, multicomponent analysis is possible. In surface enhanced Raman spectroscopy (SERS),^{1,2} Raman scattering from molecules adsorbed on roughened gold (Au), silver (Ag), and copper (Cu) surfaces can be enhanced by as much as 10^6 to 10^7 . In addition, water is a poor Raman scatterer. Consequently, the vibrational bands due to water do not interfere in the detection of the Raman/SERS peaks caused by the analyte. These attributes in aggregate make Raman and SERS attractive techniques for field detection of environmental contaminants in both the gas phase and aqueous media. Raman/SERS have applications in industrial process control as well as in materials detection/identification in agriculture, pharmaceuticals, law enforcement/first responders, geology, and archeology. Raman spectroscopy has been used to discriminate between Green Arabica and Robusta coffee beans,³ to detect counterfeit and adulterated pharmaceuticals,⁴ to identify plastics for recycling,⁵ and to evaluate the selectivity of bifunctional anion exchange resins for perchlorate.⁶ SERS has been used to detect melamine in milk products,⁷ pesticide residues on fruit,⁸ pathogens,⁹ perchlorate,¹⁰ and uranium.¹¹

Process control and real-time on-site monitoring of analytes in the field require the use of portable, robust Raman spectrometer systems. There have been significant improvements in stabilized, small, inexpensive laser sources, optical filter technology, and charge-coupled device (CCD) detectors.¹² Consequently, a number of miniaturized Raman spectrometer systems are commercially available.¹³ These include fiber optic systems such as the InPhotonics' InPhotote and InPhotote1064 portable Raman systems, Enwave Optronics' EZRaman, and B&W Tek's MiniRam. The latter two systems are available in 532 or 785 nm excitation. B&W Tek also offers a handheld Raman system, the NanoRam. Additionally, Thermo Scientific

offers TruScan, another handheld Raman system. These handheld systems are 'point-and-shoot' and are battery operated. Ultimately, the success of using Raman/SERS for field detection of environmental contaminants, industrial process control, and materials detection/identification in agriculture, pharmaceuticals, law enforcement/first responders, geology, and archeology depends upon the performance of the Raman spectrometer system. While there have been product reviews published on Raman systems,^{13,14} these reviews only provide brief descriptions of the instruments and have indicated their costs and applications. Rarely are there published studies of side-by-side comparisons evaluating the performance of these systems. In an attempt to address this need, the Raman/SERS performance of two commercially available, portable Raman systems are evaluated in this communication. The two systems are DeltaNu's Inspector Raman system and Snowy Range Instruments' Sierra Raman system.

Experimental Reagents

Sodium chromate (Aldrich), potassium chloride (Aldrich), HPLC grade water (Aldrich), naphthalene (Fluka), and HPLC/Spectrophotometric grade ethanol (Aldrich) were used as received. The thiols cysteamine (CY) hydrochloride (Fluka) and 4-(2-mercaptoethyl) pyridinium (MEP) hydrochloride (Toronto Research Chemicals) were also used as received. Aqueous solutions of sodium chromate and potassium chloride were prepared using deionized water.

Descriptions of the portable Raman systems

The Raman Solution 785 system (Detection Limit) has a f number of 2; a fixed position, 1200 grooves mm^{-1} grating (spectral range 250–1750 cm^{-1}); and a TE cooled Kodak 0400 CCD. A fiber optic sampling probe operating at 785 nm (InPhotonics, Model RPS785-12-10) was used to deliver the laser excitation to the sample and transfer the Raman emissions to the spectrometer. The excitation source was a tunable, continuous wave (CW) laser diode (Spectra Diode Laser, SDL-8630) operating at 785 nm. A tunable optical isolator (Optics for Research, Model IO-7-NIR) was used to prevent backscatter of the laser beam into



the laser cavity. The 785 nm laser line was focused into the silica/silica clad, 100 μm diameter, excitation fiber using a 5 X microscope objective lens. The spot size on the sample is therefore 100 μm in diameter.

The Inspector Raman (DeltaNu) is a handheld system. It has a 785 nm laser that can operate at 120 mW, a 1024 element, linear CCD array, dispersive spectrometer (operational range is 200–2000 cm^{-1} and resolution is less than 8 cm^{-1}), optical components, and rechargeable batteries. The spot size on the sample is about 30 μm in diameter.

The Snowy Range Instruments Sierra portable Raman system has a 785 nm laser that can operate at 100 mW, a 3000 element, linear, cooled, NIR enhanced CCD array, dispersive spectrometer (operational range is 200–3200 cm^{-1} and resolution is 10 cm^{-1}), and optical components. The Sierra is designed to be a bench-top instrument and does not have rechargeable batteries. The spot size on the sample is about 30 μm in diameter. This particular system has three-way sampling for side, bottom, and ‘point and shoot.’ It also has an ‘Orbital Raster Scan (ORS)’ technology. The instrument can raster the tightly focused laser beam over the sample. This allows interrogation of a larger sample area without loss of resolution.

Preparation of Au/MEP SERS substrates

An insulated copper wire was soldered to a 0.75 cm length of 2 mm diameter gold wire (Aldrich, 99.9%). A chemically resistant epoxy (Epoxy Patch 1C, Hysol) was used to pot the gold wire inside a 5 mm o.d. glass tube. Prior to use, the gold disk of the electrode was electrochemically roughened in a 0.1 M KCl solution using a PAR 273A potentiostat/galvanostat under computer control. The gold surface was roughened by cycling the electrode from –280 mV vs. Ag/AgCl reference electrode (holding 10 s) to 1220 mV vs. Ag/AgCl (holding 5 s) at a scan rate of 500 mV s^{-1} . This oxidation-reduction cycle (ORC) was repeated 25 times in succession. After electrochemical roughening, the gold electrode was rinsed with water (HPLC grade, Aldrich) and then with ethanol (HPLC/Spectrophotometric grade, Aldrich). The electrode was immersed in a dilute MEP solution in ethanol and allowed to react for approximately 24 h to form a self-assembled monolayer (SAM). Before use,

the Au/MEP electrode was rinsed thoroughly with ethanol and then water.

Preparation of Ag/CY SERS substrate

A 1 cm^2 area, 0.1 mm thick silver foil (Aldrich, 99.9%) was electrochemically roughened in a 0.1 M KCl solution using a PAR 273A potentiostat/galvanostat under computer control. To roughen silver, 25 successive ORCs from –300 to 1200 mV vs. the Ag/AgCl reference electrode at a sweep rate of 500 mV s^{-1} were used. For each scan, the potential was held for 1.3 s at the positive limit and 30 s at the negative limit. After electrochemical roughening, the silver foil was rinsed with water (HPLC grade, Aldrich) and then with ethanol (HPLC/Spectrophotometric grade, Aldrich). The foil was immersed in a dilute CY solution in ethanol and allowed to react for approximately 24 h to form a SAM. Before use, the Ag/CY SERS substrate was rinsed thoroughly with ethanol and then water.

Raman and SERS measurements

Raman spectra of chromate solutions were collected in cuvettes that were rigidly held in place within the focal length of the fiber optic probe of the Detection Limit system and the optics of the DeltaNu system. For the Snowy Range Instruments system, the cuvettes were placed in the sample chamber.

A schematic of the flow-through cell used to evaluate the response of Au/MEP SERS substrates is shown in Figure 1. The body of the flow-through cell is constructed of Teflon[®]. A 25 mm diameter, 2 mm thick sapphire disk was used as the optically transparent window. A Kel-F window retainer holds the window in place. An O-ring provides a leak-tight seal between the window and the cell body. A Teflon[®] bushing holds the SERS substrate in place. A second O-ring provides a leak-tight seal around the glass tube of the SERS substrate. The inlet and outlet consist of 20 gauge stainless steel tubes that have been epoxied in place using a chemically resistant epoxy (Epoxy Patch 1C, Hysol). The volume of the cell is ~0.5 mL.

Manipulation of spectral data

All manipulations of the spectral data were done using GRAMS/AI7 (ThermoGalactic). This software

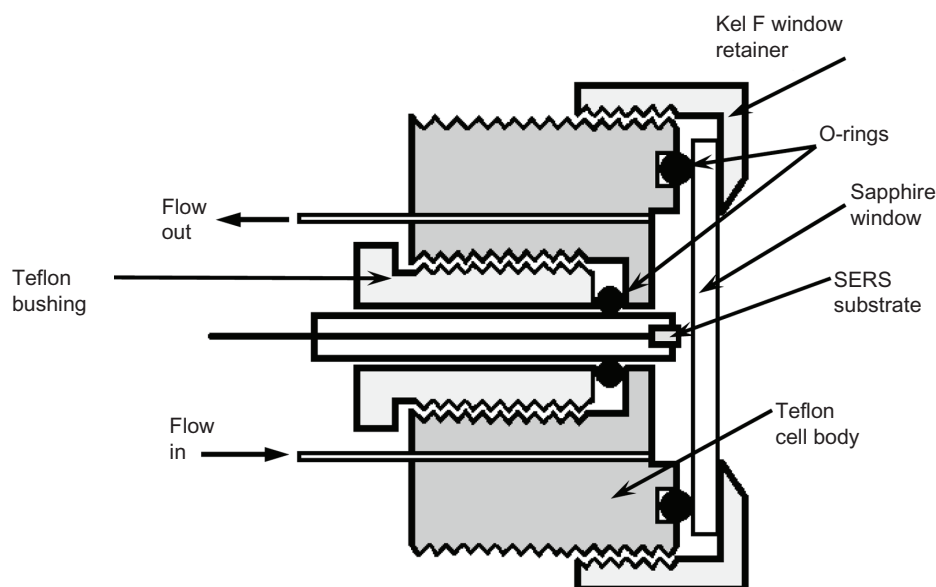


Figure 1. Schematic of the flow-through sample cell used to obtain SERS spectra as a function of analyte concentration.

package was used to do spectral subtractions and to integrate peak area.

Results and Discussion

Detection limit raman system

The Detection Limit Raman system uses an external laser as its excitation source. In this system, the laser is a tunable CW laser that requires vibration stabilization. Consequently, it is not ideal for use as a field deployable system. However, the Detection Limit Raman system was used earlier to conduct Raman and SERS laboratory studies of anions,^{15–17} detection of volatile organic compounds using a SERS substrate mounted on a thermoelectric cooler,¹⁸ and the characterization of bifunctional anion exchange resins using Raman spectroscopy.⁶ Because of its documented use in both Raman and SERS characterizations of chemical systems, the performance of this instrument is used as the benchmark for comparison of the Delta Nu Inspector Raman and Snowy Range Instruments Sierra portable Raman systems.

Figure 2A shows Raman spectra of aqueous chromate solutions and a plot of chromate peak area as a function of chromate concentration. The experimental parameters used to obtain these spectra are summarized in Table 1. The chromate concentrations of the solutions range between 500 and 10,000 ppm. The band at 847 cm^{-1} in Figure 2A(i) is actually comprised of two overlapping peaks at 850 and 894 cm^{-1}

due to the symmetric and asymmetric Cr-O stretching modes of the chromate anion.¹⁷ The plot of chromate peak area as a function of chromate concentration, Figure 2A(ii), is linear. The concentration profile was analyzed using a Marquardt¹⁹ nonlinear least-squares fitting routine to obtain the values of the slope and intercept as well as their standard deviations. The analysis also determines R^2 , the coefficient of determination. These values are tabulated in Table 2. The limit of detection (LOD) is given by the following relationship:^{20–22}

$$LOD = \frac{3\sigma}{m} \quad (1)$$

where σ is the uncertainty in the y-intercept, b , and m is the slope of the line. The LOD is also tabulated in Table 1.

Earlier Turyan and Mandler²³ reacted Au electrodes with MEP to form a SAM. They then used these Au/MEP modified electrodes to preconcentrate chromate prior to detection by cathodic stripping square wave voltammetry. The MEP coating exhibits great selectivity for hexavalent chromium. In their studies, Turyan and Mandler showed that the anions chloride, nitrate, phosphate, acetate, and perchlorate did not interfere in the detection of Cr(VI). Figure 3A(i) shows SERS spectra of a Au substrate coated with MEP as a function of chromate concentration. These

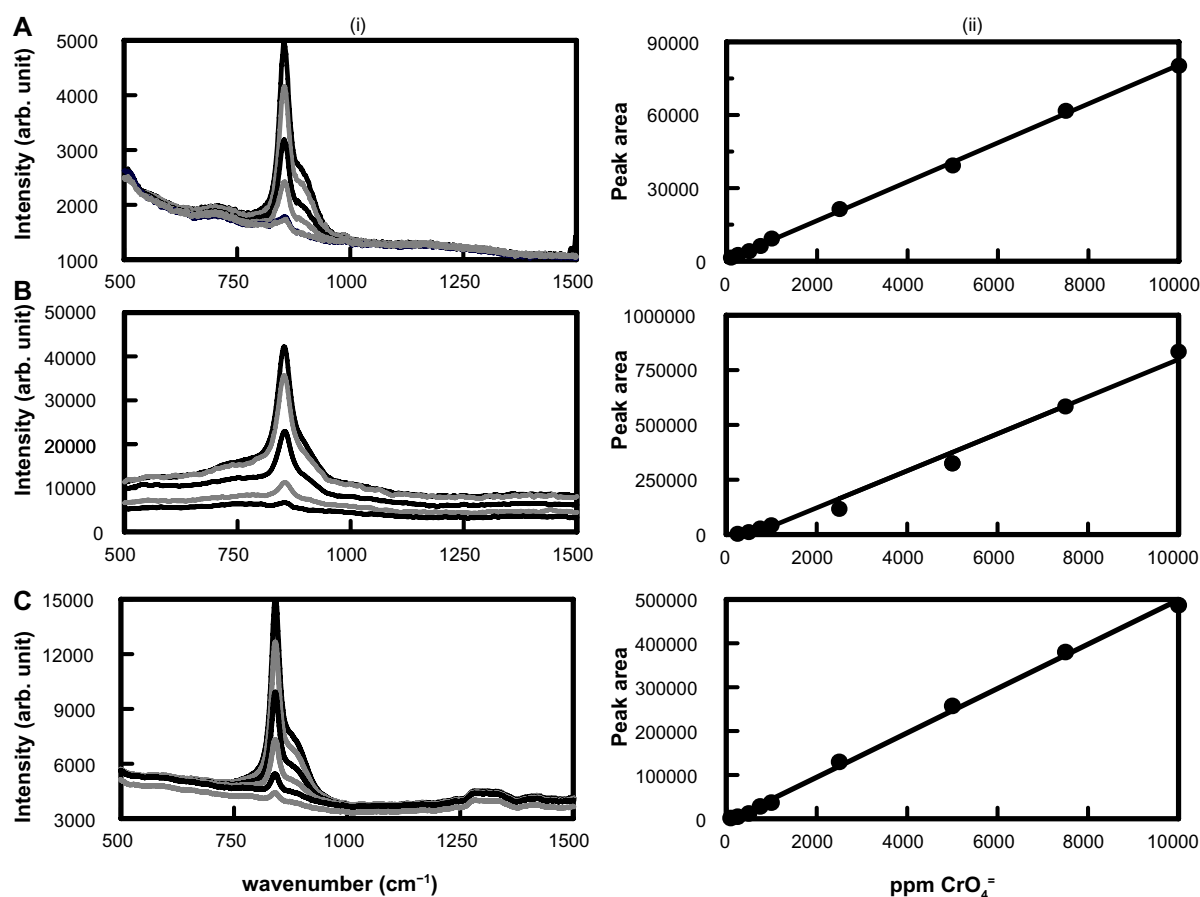


Figure 2. Raman spectra (i) and concentration response (ii) measured for chromate solutions for (A) the Detection Limit, (B) DeltaNu, and (C) Snowy Range Mountain Raman systems. For (A) the spectra shown are for 500, 750, 2500, 5000, 7500, and 10000 ppm chromate solutions. For (B) the spectra shown are for 750, 2500, 5000, 7500, and 10000 ppm chromate solutions. For (C) the spectra shown are for 500, 750, 2500, 5000, 7500, and 10000 ppm chromate solutions. Experimental parameters used to obtain the spectra are summarized in Table 1.

spectra were obtained using the Detection Limit Raman system. The experimental parameters used to obtain these spectra are summarized in Table 3. As the chromate concentration increases, a peak due to the symmetric and asymmetric Cr-O stretching modes of chromate anion grows in at $\sim 835\text{ cm}^{-1}$. The chromate peak overlaps with peaks due to the MEP coating. To determine the chromate peak area, the spectral contributions of the coating must be subtracted out. Once the subtraction is done, integration under the resultant peak results in the peak area. Figure 3A(ii) shows examples of some of these difference spectra.

The broad band due to the chromate anion is identified in Figure 3A(ii). The difference spectra in Figure 3A(ii) also show increased intensity of the MEP peaks at $1100\text{--}1200\text{ cm}^{-1}$ and $1450\text{--}1630\text{ cm}^{-1}$. These peaks are primarily assigned to the pyridine ring vibrational modes.¹⁷ The intensity changes indicate that the pyridine ring is involved in the complexation reaction and that hydrogen bonding was occurring between the chromate and the MEP. Molecular modeling of the SAM and chromate showed the presence of microcavities between adjacent MEP moieties on the SERS surface with a three-dimensional structure

Table 1. Experimental parameters used to obtain the spectra shown in Figure 2.

Instrument	Spot size (μm)	Laser excitation (nm)	Laser power (mW)	Acquisition time	Resolution setting
Detection Limit	100	785	62	Avg. 5 @ 20s	–
Inspector Raman	30	785	75	Avg. 10 @ 10s	low
Sierra	30	785	80	Avg. 10 @ 5s	–

Table 2. Summary of LODs, R^2 , and the values of m and b that describe the concentration responses shown in Figure 2.^a

Instrument	m	b	R^2	LOD (ppm)
Detection Limit	7.77 ± 0.08	640 ± 360	0.9993	139
Inspector Raman	84.6 ± 3.6	-47000 ± 8000	0.9891	284
Sierra	50.53 ± 0.86	-6300 ± 3900	0.9980	231

Note: ^aThe concentration response is described by: $y = mx + b$ where y is the chromate peak area and x is ppm chromate.

complementary in both shape and chemical functionality to that of the chromate anion.¹⁷

A plot of chromate peak area as a function of chromate concentration is shown in Figure 4A. This is the calibration curve. At low chromate concentration, the chromate peak area increases linearly with concentration. At higher chromate solution concentrations, the response levels off as the adsorption sites of the substrate become fully occupied. It was found that this calibration curve is adequately described by the following numerical equation:

$$y = c + a(1 - \exp(-bx^{0.2})) \quad (2)$$

where y is chromate peak area and x is chromate concentration in ppm. The concentration profile was analyzed using a Marquardt¹⁹ nonlinear least-squares fitting routine to obtain the values of a , b , c , as well as their standard deviations. The analysis also determines R^2 . These values are tabulated in Table 4.

The SERS substrates used in this investigation were prepared by electrochemically etching the Au or Ag surface. It is well known that the magnitude of the SERS response is determined by the morphological features of the substrate, ie, the size and shape of the dendrites formed during the etching process. Because of the manner in which the SERS substrate was prepared (a 3 mm diameter Au wire potted inside a glass tube), we were unable to characterize these features. However, etching parameters were developed which gave reproducible substrates showing minimal variability. These etching parameters were used to prepare all the Au/MEP SERS active substrates used in this investigation.

DeltaNu inspector Raman system

Unlike the Detection Limit system, the DeltaNu system software has a background correction feature. The SERS spectrum for Au/MEP, Figure 5A, shows SERS peaks due to the MEP SAM and chromate on a broad

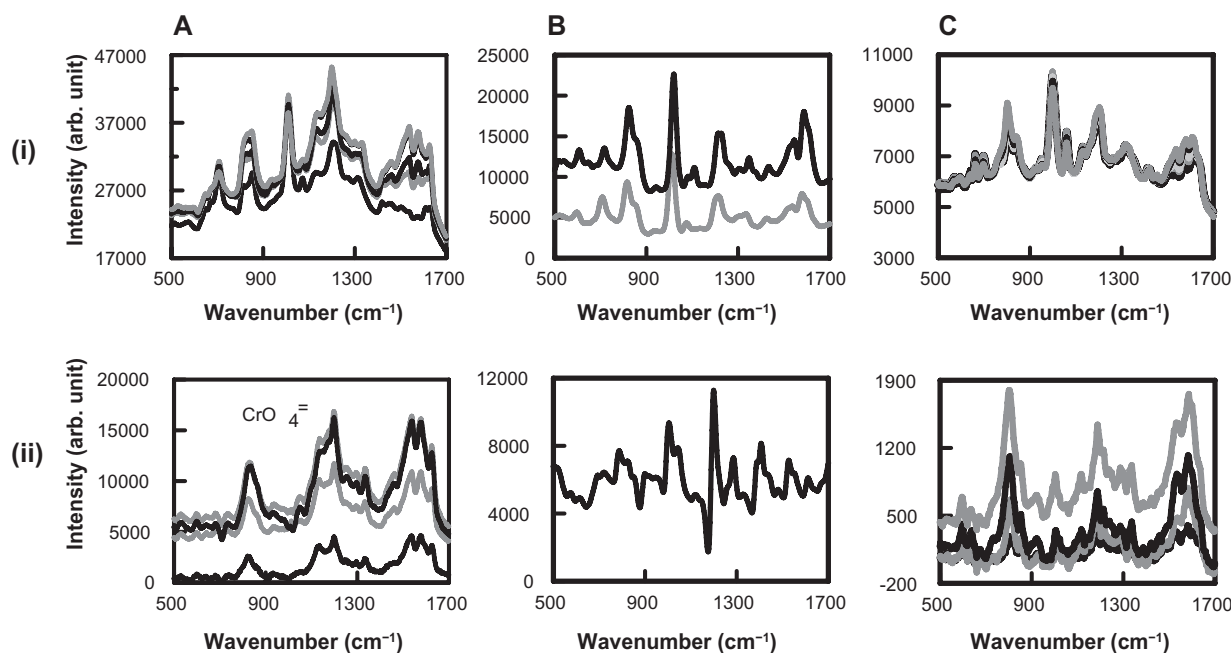


Figure 3. SERS spectra (i) and difference spectra (ii) of Au/MEP measured for chromate solutions and (A) the Detection Limit, (B) DeltaNu, and (C) Snowy Range Mountain Raman systems. For (A) the spectra shown are for 0, 2.5, 25, 2500, and 10000 ppm chromate solutions. For (B) the background corrected spectra shown are for 0 (gray) and 10000 (black) ppm chromate solutions. For (C) the spectra shown (bottom to top) are for 0, 25, 100, 500, 1000, and 5000 ppm chromate solutions. Experimental parameters used to obtain the spectra are summarized in Table 3.

Table 3. Experimental parameters used to obtain the spectra shown in Figure 3.

Instrument	Spot size (μm)	Laser excitation (nm)	Laser power (mW)	Acquisition time	Resolution setting
Detection Limit	100	785	80.2	Avg. 5 @ 20s	–
Inspector Raman	30	785	34	Avg. 10 @ 5s	Low
Sierra	30	785	30	Avg. 10 @ 2s	–

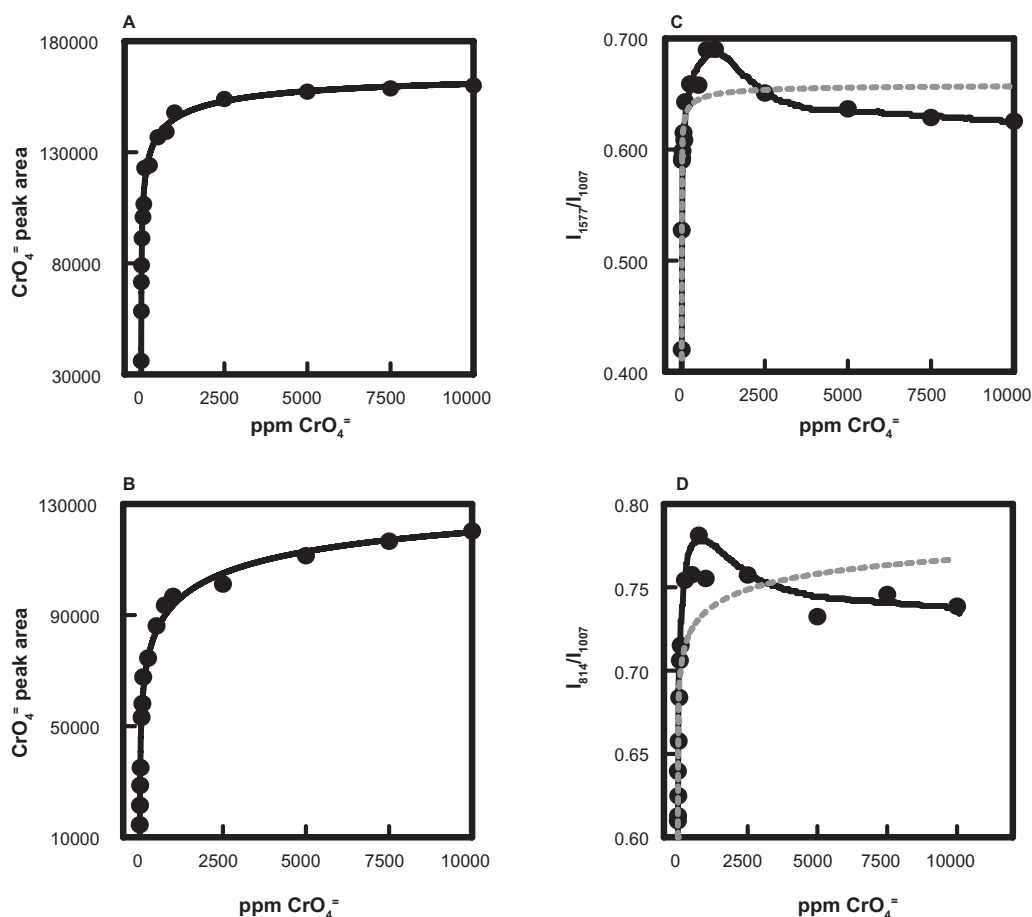


Figure 4. The Au/MEP-chromate calibration curves measured using the (A) Detection Limit, (B) Snowy Range Mountain, and (C and D) DeltaNu Raman systems. For (A) and (B) chromate peak area is plotted as a function of chromate concentration. The line going through the points was calculated using the equation $y = c + a(1 - \exp(-bx^{0.2}))$ where the values of a , b , and c are tabulated in Table 4. For (C) and (D) the peak intensities at 1577 and 814 cm^{-1} are ratioed to the 1007 cm^{-1} peak. These ratios are plotted as a function of chromate concentration. The black, solid lines are drawn through the points. The gray, dotted lines were calculated using the equation $y = c + a(1 - \exp(-bx^{0.2}))$ where the values of a , b , and c are tabulated in Table 4.

Table 4. Summary of the values of a , b , and c that describe the Au/MEP-chromate concentration responses shown in Figure 4 and R^2 .^a

Instrument	c	a	b	R^2
Detection Limit	-60000 ± 12000	226000 ± 10000	0.597 ± 0.046	0.9916
Sierra	-47100 ± 9400	188100 ± 6000	0.346 ± 0.047	0.9878
Inspector Raman	0.412 ± 0.024	0.246 ± 0.025	0.85 ± 0.16	0.8850
I_{1577}/I_{1007}	0.591 ± 0.032	0.207 ± 0.055	0.30 ± 0.20	0.6806
I_{814}/I_{1007}				

Note: ^aThe concentration response is described by: $y = c + a(1 - \exp(-bx^{0.2}))$ where y is either the chromate peak area or the ratio of peak intensities and x is ppm chromate.

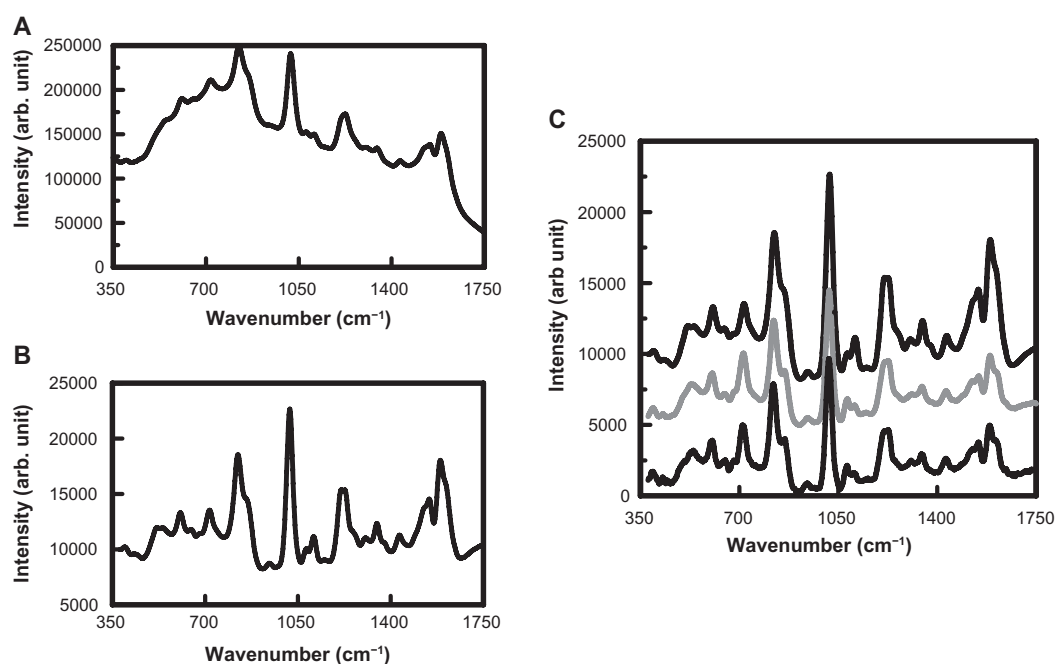


Figure 5. Au/MEP SERS spectra obtained using the Delta Nu Inspector Raman system. Spectra obtained using 785 nm excitation at 34 mW and averaging ten 5 s spectra. The chromate concentration is 10000 ppm. (A and B) Uncorrected and background corrected SERS spectra respectively. (C) Spectra obtained as a function of spectral resolution where, top to bottom, the resolution is low, medium, and high.

SERS continuum. The DeltaNu software filters in the Fourier domain to remove the broad background.²⁴ Broad band features appear at low frequencies in the Fourier domain. A high pass filter is used to reject broad bands. The results of the background correction feature for the SERS spectrum is shown in Figure 5B. It can be seen with the Raman/SERS, that there is a significant decrease in the signal intensity. The peaks are also narrowed. In Figure 5A, the full width at half maximum (FWHM) for peaks at 819 and 1007 cm^{-1} are 76.2 and 38.1 cm^{-1} , respectively. After background correction, the FWHM for peaks at 819 and 1007 cm^{-1} are 47.6 and 19 cm^{-1} , respectively.

Another feature of the DeltaNu system not present in the Detection Limit system is three possible resolution settings. The resolution is changed by conducting a Fourier Transform Apodization.²⁵ SERS spectra obtained using these resolution settings are shown in Figure 5C. It can be seen that the low resolution setting shows the highest signal. With the medium and high resolution settings, better separation of the peaks between 770–900 cm^{-1} and 1475–1665 cm^{-1} is observed.

Raman spectra of chromate solutions and SERS spectra of Au/MEP in the presence of chromate were obtained using the DeltaNu system to compare its performance with that of the Detection Limit system.

Figure 2B shows Raman spectra of aqueous chromate solutions and a plot of chromate peak area as a function of chromate concentration. The chromate concentrations of the solutions range between 750 and 10,000 ppm. The experimental parameters used to obtain these spectra are summarized in Table 1. The spectra were obtained with the resolution on low to increase the sensitivity and without background correction. Compared to the spectra obtained using the Detection Limit system, Figure 2A(i), the resolution is decreased. In the spectra shown in Figure 2A(i), it can be clearly seen that the band at 847 cm^{-1} is comprised of two overlapping peaks at 850 and 894 cm^{-1} . In Figure 2B(i), only a slight shoulder on the high wavelength side is observed in the band at 847 cm^{-1} , suggesting that this band is actually two overlapping peaks. The plot of chromate peak area as a function of chromate peak area, Figure 2B(ii), is linear. The concentration profile was analyzed using a Marquardt¹⁹ nonlinear least-squares fitting routine. Table 2 tabulates the values of the slope, intercept, R^2 , and LOD. The R^2 is a little lower than that obtained with the Detection Limit system, indicating that there is a little more scatter in the data. This scatter results in a higher LOD than that obtained using the Detection Limit system.

In Figure S1 (see Supplementary material), SERS spectra of a Au substrate coated with MEP are shown as a function of chromate concentration. These spectra were obtained with the background correction feature disabled. Figure S1B shows difference spectra obtained by subtracting the spectral contributions of the Au/MEP SAM from the Au/MEP-chromate spectra. The difference spectra show increased intensity of the MEP peaks at 1180–1275 cm^{-1} and 1400–1600 cm^{-1} . The increase in the peak intensities indicates that the chromate ion is interacting with the MEP SAM. However, the difference spectra show no peaks due to chromate. This indicates that the SERS broad continuum masks the chromate peak. This was not observed in the difference spectra obtained using the Detection Limit system.

Results obtained for a Au/MEP substrate as a function of chromate concentration and with the background correction feature activated are shown in Figure S2 (see Supplementary material). The spectral peaks are narrow but the intensity of the 1007 cm^{-1} peak varies considerably. This contrasts with the results obtained for the Detection Limit system results shown in Figure 3A and the background correction feature disabled results shown in Figure S1 in the Supplementary material, which show no variation in peak intensities. This variability in peak intensity is probably an artifact caused by the background correction algorithm.

Figure 3B(i) shows spectra obtained for the Au/MEP SAM in the presence and absence of 10,000 ppm chromate. Besides the variability in intensity of the peak at 1007 cm^{-1} , it can be seen that the peaks do not line up, indicating that shifts in the peak positions have occurred. The resultant difference spectrum, Figure 3B(ii), shows that it is not possible to obtain a clean chromate peak whose peak area can be measured and a calibration curve generated. Another way to generate a calibration curve is to ratio MEP peaks which change with chromate concentration with one that does not change. The Detection Limit spectral results summarized in Figure 2A(i) show that the large peak at $\sim 1007 \text{ cm}^{-1}$ does not vary with increasing chromate concentration, whereas the peaks at $\sim 814 \text{ cm}^{-1}$ (due to overlapping chromate and MEP peaks) and 1577 cm^{-1} do. The intensities of these peaks for the DeltaNu spectra, Figure S2 (see Supplementary material)

were measured. Plots of the 1577 and 814 cm^{-1} peaks ratioed to 1007 cm^{-1} peak as a function of chromate concentration are shown in Figure 4C and D, respectively. The two plots are very similar. Between 0–1000 ppm chromate the response is linear. It maximizes at 1000 ppm, falls off, and then levels off. This deviates from the plot obtained using the Detection Limit system shown in Figure 4A and the curves are not described by equation 2. In the spectral regions near 1577 and 814 cm^{-1} , there are overlapping MEP and chromate peaks. The deviation from the Detection Limit calibration curve could be due to intensity variations in these overlapping MEP and chromate peaks as a function of chromate concentration.

Snowy Range Instruments Sierra Raman system

Like the DeltaNu system, the software of the Snowy Range Instruments system has a background correction feature. It is an automated version of the multipoint baseline feature used in GRAMS.²⁵ The algorithm first finds all of the valleys in the spectrum. It then creates a ‘baseline’ spectrum by creating lines from valley to valley. This ‘baseline’ spectrum is then subtracted from the original. The use of this background feature is summarized in Figure 6A. The top spectrum in black is the one measured while the ‘baseline’ spectrum is in gray. The bottom spectrum is the resultant baseline-corrected spectrum. It can be seen that the broad SERS continuum has been removed. It can also be seen that, unlike the DeltaNu background correction algorithm, there is no substantial decrease in line intensity and no shifts in the spectral peaks. The Snowy Range Instruments system also has a smoothing feature that is based upon the Savitky-Golay method.²⁵ Figure 6B shows the results of using both the background and smoothing features. The top spectrum is the measured spectrum and the bottom is the background-corrected, smoothed spectrum.

A unique feature of the Snowy Instruments portable Raman spectrometer is its rastering capability. All other portable Raman systems focus the excitation laser beam on a single, fixed spot on the sample surface. In these fixed spot systems, there is a trade-off between resolution and sensitivity. Raman peaks are used to identify a chemical compound by their characteristic Raman signature. Some of these

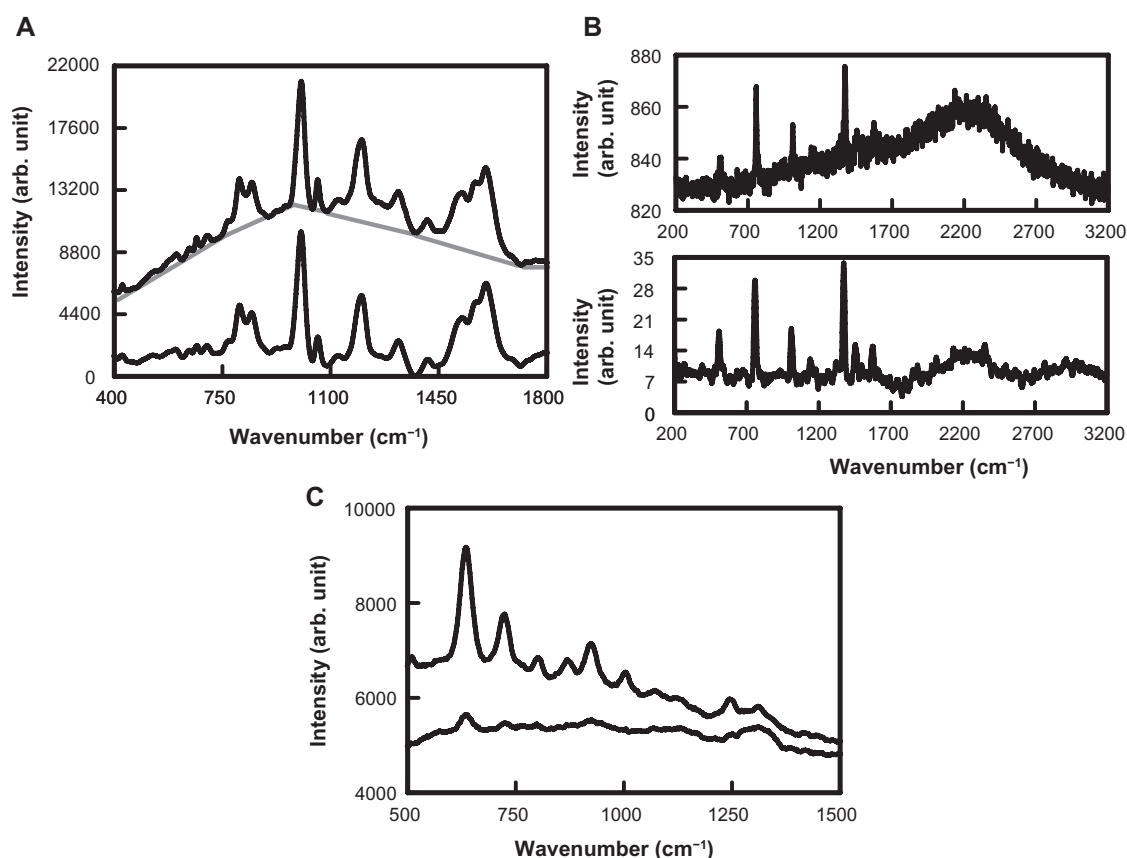


Figure 6. (A) SERS spectra obtained using the Snowy Range Instruments Raman system where the top and bottom spectra are the Au/MEP and 2500 ppm chromate uncorrected and background corrected, respectively. The gray line in the top spectrum is the 'baseline' spectrum. Spectra were obtained using 785 nm excitation at 80 mW and averaging thirty 1 s spectra. (B) Raman spectra of naphthalene obtained with the background correction and smoothing features deactivated (top spectrum) and activated (bottom spectrum). Spectrum was obtained using 785 nm excitation at 0.03 mW power and 0.5 s acquisition time. (C) SERS spectra of Ag/CY obtained with (top) and without (bottom) rastering. Spectra were obtained using 785 nm excitation at 80 mW and averaging ten 3 s spectra.

commercially available, fixed spot Raman systems interrogate a small spot size (typically on the order of 50 microns or less) in order to resolve the Raman peaks. However, a small spot size will result in a less intense signal as only a small area of the sample is interrogated. To overcome sensitivity issues, these small spot, fixed point systems typically use higher laser powers. However, these high laser powers can cause the sample to burn, resulting in changes to the Raman/SERS spectra. This is shown in the Au/MEP spectra shown in Figure S3A (see Supplementary material). As a result, the chemical compounds making up the sample become degraded and the chemical species cannot be positively identified.

Other commercially available, fixed spot Raman systems interrogate a large spot size (100 microns or larger). A larger spot size will cover more of the sample surface. This will increase sensitivity and will not cause the sample to burn. However, it will result

in broader peaks which can impact sample identification. The rastering capability of the Snowy Range Mountain system overcomes the problems experienced with fixed spot Raman systems. The small spot size laser (30 microns) is rapidly scanned (rastered) over a larger sample area (typically 20 mm²). A small spot size assures high resolution. Higher laser powers can be used because the same spot on the surface is not interrogated. In addition, the rastering capability is particularly useful for SERS, but only if the SERS substrate is fairly large. On a SERS substrate, there are 'hot spots' which exhibit greatly enhanced signals. A fixed spot Raman system interrogates a limited number of these SERS active hotspots and the resulting spectrum is overwhelmed by the background caused by the surrounding substrate. Rastering allows one to quickly scan a large area and interrogate more SERS active hot spots. This sampling approach collectively reduces the background of the spectrum by averaging

the SERS active hotspots, which have a more intense Raman signature than the interference from the surrounding substrate. This greatly increases both sensitivity and resolution. To demonstrate this, Figure 6C shows SERS spectra obtained for a Ag/CY substrate with and without rastering. The SERS substrate had a surface area of $\sim 1 \text{ cm}^2$. Both spectra were obtained using the same laser power (80 mW) and acquisition time. The SERS signal due to the CY coating obtained without rastering has low intensity. The signals due to the C-S stretching modes of the gauche and trans conformers of CY, between $600\text{--}750 \text{ cm}^{-1}$, are barely discernible. Peaks due to the CH_2 and NH_3 vibrational modes are not discernible above the SERS continuum. At this laser power, the SERS substrate is becoming damaged. With rastering, the same spot is not interrogated, which minimizes damage to the SERS substrate, and more 'hot spots' are sampled. As a result, not only are the peaks due to the C-S stretching modes observed in the resultant spectrum, but peaks due to the CH_2 and NH_3 vibrational modes are also clearly observed in the spectrum.

Raman spectra of chromate solutions and SERS spectra of Au/MEP in the presence of chromate were obtained using the Snowy Range Mountain system to compare its performance with that of the Detection Limit system. Figure 2C(i) shows Raman spectra of aqueous chromate solutions obtained using the Snowy Range system. The chromate concentrations of the solutions range between 500 and 10,000 ppm. The shape of the chromate peaks are very similar to those obtained for the Detection Limit system, Figure 2A(i). Figure 2C(ii) is a plot of chromate peak area as a function of chromate peak area. The resultant plot is linear. The concentration profile was analyzed using a Marquardt¹⁹ nonlinear least-squares fitting routine. Table 2 tabulates the values of the slope, intercept, R^2 , and LOD. The R^2 is similar to that obtained with the Detection Limit system. The LOD is higher than that obtained using the Detection Limit system but lower than that for the DeltaNu Inspector Raman.

SERS spectra of Au/MEP substrates as a function of chromate concentration were obtained using laser powers of 80 and 30 mW. The spectral data and concentration plots obtained using the higher laser power are shown in Figure S3 (see Supplementary material). The intensities of the peaks over time were observed to decrease in the SERS spectra shown in Figure S3A,

indicating that the SERS substrate is degrading. This was not observed in the spectra obtained using the Detection Limit system, Figure 3A(i). A significant difference between these Raman systems is that the Detection Limit system used fiber optics while the Snowy Range Raman system does not. Consequently, the spot size on the sample using the Detection Raman system was $100 \mu\text{m}$. The spot size for the Snowy Range Raman system is about $30 \mu\text{m}$ in diameter. As a result, the laser power density at the sample is more than an order of magnitude greater for the Sierra Raman systems than it was for the Detection Raman system. Using a laser power of 30 mW, spectra were obtained for a conventional Au/MEP SERS substrate as a function of chromate concentration using the Sierra Raman system. These spectra are shown in Figure 3C(i). With a lower laser power, no variation in signal intensity of the 1007 cm^{-1} peak was observed and the spectra were similar to those obtained using the original Detection Limit Raman system; Figure 3A(i). Figure 3C(ii) shows the difference spectra. The resolution of the peaks is comparable to what was obtained using the Detection Limit system, Figure 3A(ii). The resultant calibration curve for Au/MEP and chromate, Figure 4B, was also similar to that obtained using the original Detection Limit Raman system. The calibration curve is adequately described by equation 2. The concentration profile was analyzed using a Marquardt¹⁹ nonlinear least-squares fitting routine to obtain the values of a , b , c , as well as their standard deviations. The analysis also determines R^2 . These values are tabulated in Table 4.

Conclusions

In this communication, the performances of the DeltaNu Inspector Raman and the Snowy Range Instruments Sierra Raman systems were evaluated. The spectral results obtained using these systems were compared with those obtained with the Detection Limit Raman Solution 785 system. The Detection Limit system was used as the benchmark for comparison as it had been used previously to characterize chemical systems by both Raman spectroscopy and SERS.

Both the DeltaNu and Snowy Range Instruments Raman systems are portable, complete systems comprising of a laser operating at 785 nm, linear CCD array, optical filters, dispersive element, lenses, and other optical components. While the DeltaNu system



has rechargeable batteries, the Snowy Range Instruments system does not. Both systems were used to obtain Raman and SERS spectra.

For Raman spectroscopy, the DeltaNu and Snowy Range Instruments Raman systems gave results comparable to that obtained for the Detection Limit system. With regards to SERS, there were significant differences between the two portable systems. The DeltaNu system has a background correction feature based upon filtering in the Fourier domain. When SERS spectra of the Au/MEP substrates as a function of chromate concentration were obtained with the background correction feature deactivated, changes in the peaks due to MEP were observed indicating that chromate was interacting with the coating. However, when spectral subtractions were done, no peaks due to chromate were observed, indicating that the chromate peak was masked by the broad SERS continuum. With the background correction feature activated, the peaks were narrower. However, variations in intensity and peak position were observed, making it impossible to do spectral subtractions. A calibration curve could be generated by ratioing MEP peaks that changed upon complexation with chromate to a MEP peak that did not change. In contrast, the SERS spectra obtained with the Snowy Range Instruments system were similar to those obtained using the Detection Limit system. Due to the smaller spot size, spectra must be obtained at a lower laser power to prevent burning of the coating. The Snowy Range Instruments system also has a background correction feature that is essentially an automated multipoint baseline correction used by GRAMS. Consequently, no variations in peak intensity or peak position are observed using this background correction feature. The Snowy Range Instruments system also has a smoothing feature and has the capability to raster the laser light over the sample surface.

In conclusion, both the DeltaNu and Snowy Range Instruments Raman systems are adequate for applications relying on Raman spectroscopy. However, for applications relying on SERS, the Snowy Range Instruments Raman system performs better than the DeltaNu system.

Author Contributions

Conceived and designed the experiments: PAMB, MDP. Analyzed the data: PAMB, MDP. Wrote the

first draft of the manuscript: PAMB, MDP. Contributed to the writing of the manuscript: PAMB, MDP. Agree with manuscript results and conclusions: PAMB, MDP. Jointly developed the structure and arguments for the paper: PAMB, MDP. Made critical revisions and approved final version: PAMB, MDP. All authors reviewed and approved of the final manuscript.

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Competing Interests

Author(s) disclose no potential conflicts of interest.

Disclosures and Ethics

As a requirement of publication the authors have provided signed confirmation of their compliance with ethical and legal obligations including but not limited to compliance with ICMJE authorship and competing interests guidelines, that the article is neither under consideration for publication nor published elsewhere, of their compliance with legal and ethical guidelines concerning human and animal research participants (if applicable), and that permission has been obtained for reproduction of any copyrighted material. This article was subject to blind, independent, expert peer review. The reviewers reported no competing interests.

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Supplementary Figures

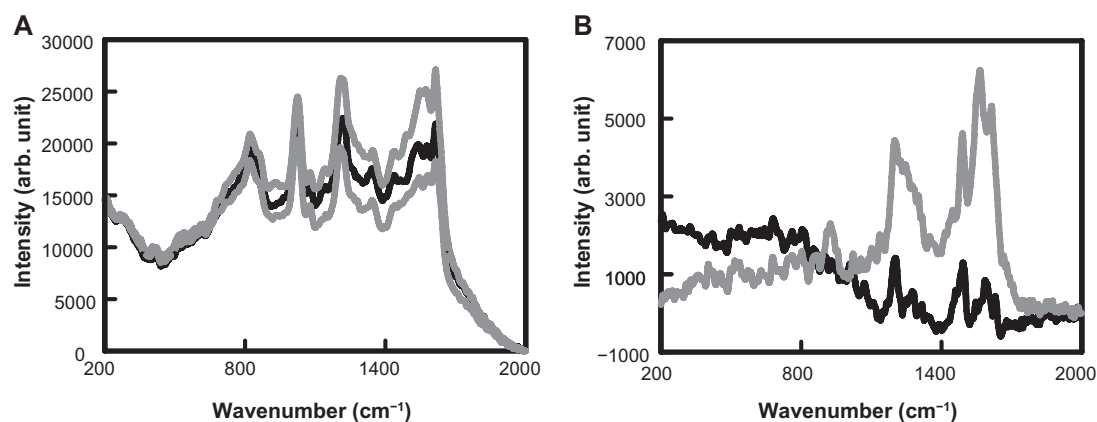


Figure S1. (A) SERS spectra of Au/MEP and, top to bottom, 5, 1, and 0 ppm chromate obtained using the using the DeltaNu Inspector Raman system. Spectra were obtained using 785 nm excitation at 34 mW, averaging ten 1 s spectra, resolution on low, and with no background correction. (B) Difference spectra for 5 (gray) and 1 (black) ppm chromate Au/MEP spectra.

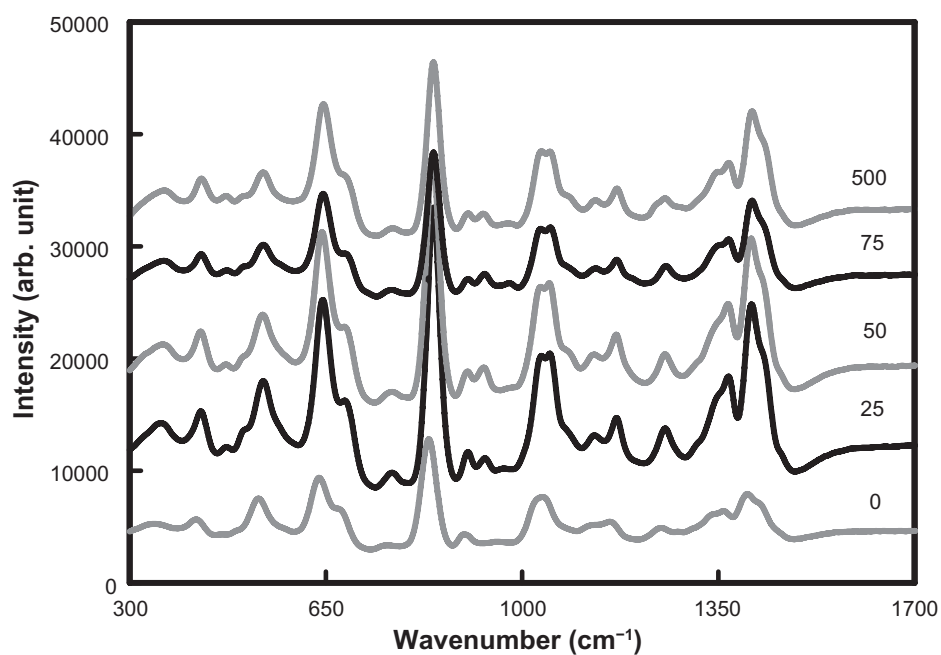


Figure S2. SERS spectra of Au/MEP obtained as a function of chromate concentration using the DeltaNu Inspector Raman system. Chromate concentration in ppm are indicated. Spectra were obtained using 785 nm excitation at 34 mW, averaging ten 5 s spectra, resolution on low, and with background correction.

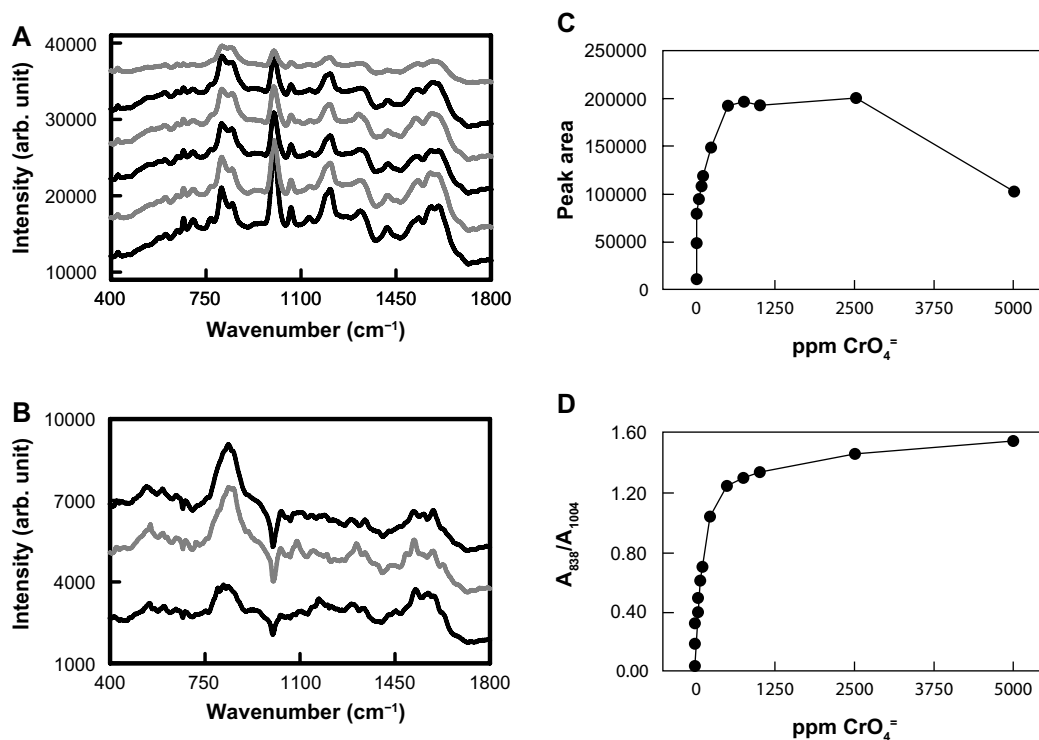


Figure S3. (A) SERS spectra of Au/MEP obtained as a function of chromate concentration using the Snowy Range Instruments Raman system. Chromate concentration in ppm are (bottom to top) 0, 25, 100, 500, 1000, and 5000 ppm. Spectra were obtained using 785 nm excitation at 80 mW and averaging thirty 1 s spectra. (B) Difference spectra obtained by subtracting the 0 ppm spectrum from the 25, 500, and 1000 ppm chromate Au/MEP spectra. (C) Plot of chromate peak area as a function of chromate concentration. (D) Concentration response for Au/MEP as a function of chromate concentration where the 838 cm^{-1} chromate peak area is ratioed to the 1007 cm^{-1} MEP peak area.