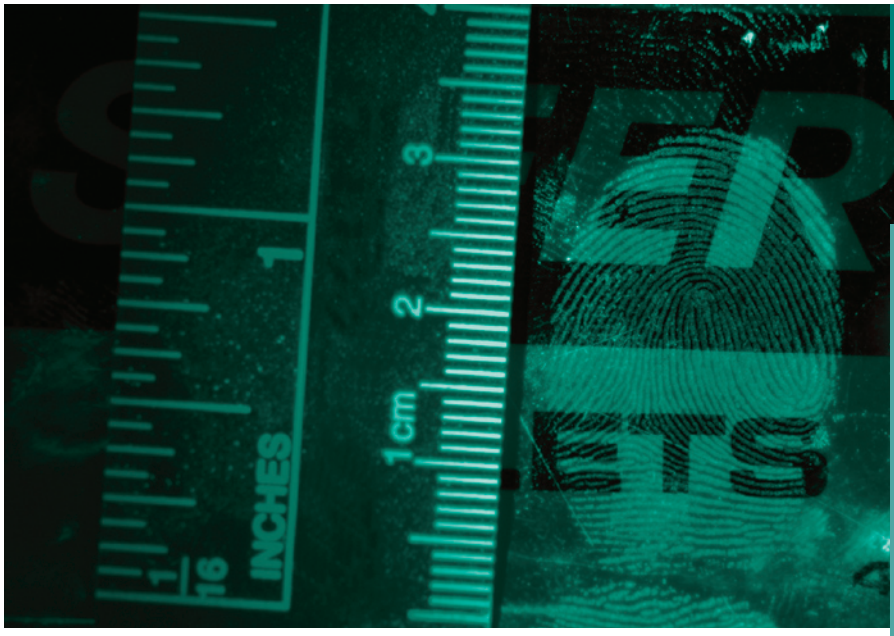


APPLICATION NOTE

Surface Enhanced Raman Spectroscopy

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# Gold Nanoparticles for SERS in Fingermark Identification

## Abstract

This note demonstrates the power of the PerkinElmer RamanStation 400 in combination with a SERS methodology to detect fingermarks on a number of substrates.

## Introduction

Fingerprint examination is a well-established method of identification using the unique arrangement of ridges on the palms of the hands. The pattern is transferred to a surface via a matrix commonly composed of secretions from glands on the fingertips. These endogenous compounds include water, lipids, fatty acids, amino acids and vitamins,[1] and leave a transparent or 'latent' fingermark. Other exogenous/contaminant substances can contribute if the hand has also been in contact with them, e.g. cosmetics, drugs, or if they have been metabolized and secreted, e.g. nicotine, medication.

Raman spectroscopy is a non-destructive technique, however, without using a method of signal enhancement, sensitivity to fingermark deposits, particularly those with endogenous components only, is a problem. Surface Enhanced Raman Spectroscopy (SERS) can provide that enhancement and the use of commercially available gold nanoparticles helps to avoid the problems of reproducibility identified with other metals, such as silver. The results presented here use SERS to image fingermark ridges composed of endogenous compounds to provide clear patterns with identifiable features.

The aims of this study were i) to determine the optimum particle size/concentration of gold nanoparticles, ii) to determine whether the use of salt aggregates is beneficial, iii) to determine whether deposition order affects the quality of the results and iv) to investigate a range of deposition surfaces.

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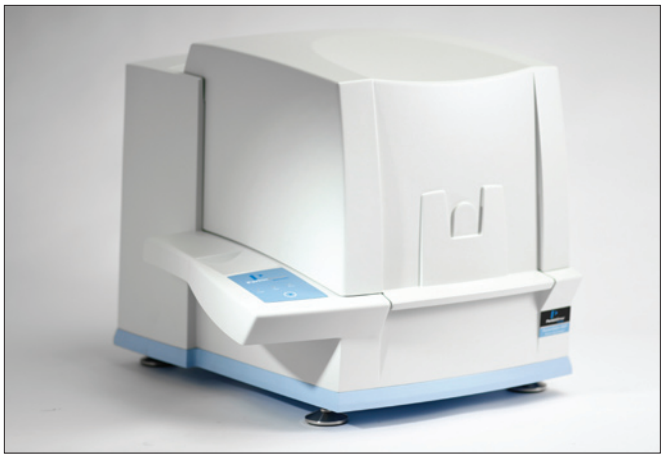

*Figure 1.* Raman spectra were acquired using a PerkinElmer RamanStation 400 benchtop Raman spectrometer.

## Experimental

### Materials and Instrumentation

Gold colloids (50 nm, 100 nm and 150 nm) were purchased from PerkinElmer. Klarite™ was obtained from D3 Technologies® Ltd (Glasgow, UK). Anhydrous sodium sulphate and acetone were purchased from Fisher Scientific® (Loughborough, UK). ALUGRAM® SIL G/UV254 pre-coated TLC aluminium plates were purchased from Sigma Aldrich® (Poole, UK). MALDI inserts were obtained from Applied Biosystems® (Foster City, CA, USA).

Raman spectra were acquired using a PerkinElmer® RamanStation™ 400 benchtop Raman spectrometer. The excitation source was a near-infrared 785 nm laser (100 mW at the sample), with a spot size of 100 µm. A spectral range of 220-3200 $cm^{-1}$ was employed. The detector was a temperature controlled Charged Coupled Device (CCD) detector (-50 °C) incorporating a 1024 x 256 pixel sensor. Spectra were acquired using Spectrum™ software and images were acquired using SpectrumIMAGE™ software, both supplied by PerkinElmer (Bucks, UK).

## Methods

### Fingerprint Deposition

Groomed latent fingermarks were prepared by rubbing the fingers on the chin, nose and forehead, five times, to yield a sebum-rich mark. Fingermarks were laid onto ALUGRAM® SIL G/UV254 pre-coated TLC aluminium plates upon removal of the silica with acetone, stainless steel MALDI inserts and Klarite™ (pre-prepared gold SERS substrate).

### Colloid Deposition

For spot deposition, 1 µL of each gold colloid (50 nm, 100 nm and 150 nm) was pipetted onto the fingermarks. For layered deposition, 100 nm colloid was applied to fingermarks laid on aluminium plates and spotless MALDI inserts in successive layers, either by spotting or spraying with an aerosol spray bottle. Each spot/layer was allowed to dry before application of the next spot/layer.

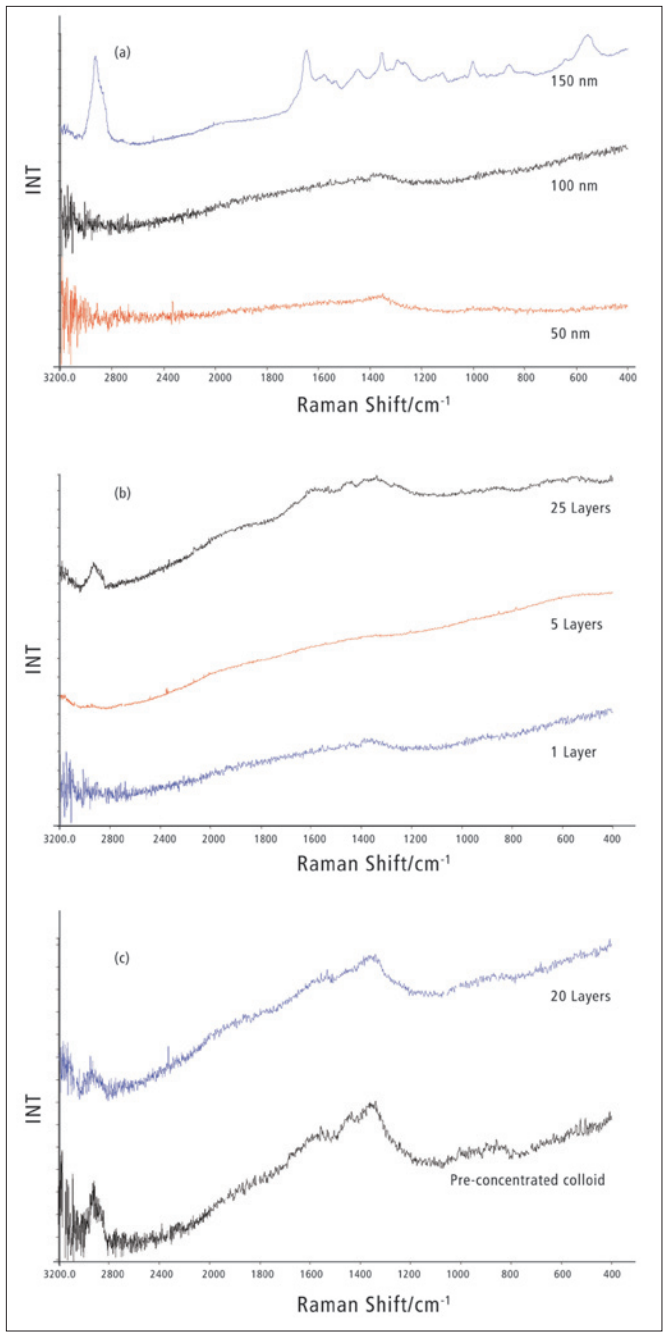


*Figure 2.* Effect of gold particle size and concentration on fingermark spectra. (a) 50 nm and 100 nm dilute colloids and 150 nm Raman specification colloid, (b) single, 5 and 25 layers of 100 nm colloid and (c) x20 pre-concentrated 100 nm colloid compared to 20 successive layers of 100 nm colloid. Fingermarks were deposited on spotless MALDI inserts and the colloid deposited on top.

### Colloid Pre-concentration

The 100 nm aqueous colloid was pre-concentrated by centrifuging 10 mL at 2750 rpm for 30 minutes and then removing 9.5 mL of the supernatant, giving a 20 times concentrated SERS suspension.

### Salt Aggregates

100 nm dilute gold colloid was mixed with a 0.1 M solution of sodium sulphate in a 1:1 ratio, to give a final concentration of 0.05 M sodium sulphate. The resulting solution was applied on top of groomed fingermarks.

### Images

For images obtained on aluminium plates and MALDI inserts, 150 nm colloid was applied directly onto the surface before groomed fingermarks were laid on top. For images on Klarite™, groomed fingermarks were laid directly onto the Klarite™ active area. Images were aquired at 0.05 mm resolution, with 2.00 s x 2 scans at each point.

## Results And Discussion

### Particle Size

Three concentrations of gold colloid were used for this study. The 150 nm colloid was specifically developed for SERS application, whereas the 50 nm and the 100 nm colloids were more dilute and intended for biological assay applications. Raman specification colloid is much more expensive than biological specification, therefore, work was carried out to investigate whether or not costs could be reduced by using a less expensive colloid. Figure 2 shows the results obtained for a fingerprint deposited on MALDI inserts; (a) illustrates the difference between the Raman and dilute colloids; (b) gives spectra for multiple layers of 100 nm colloid and (c) compares multiple layers of 100 nm colloid to pre-concentration of 100 nm colloid. As would be expected, the 150 nm Raman specification colloid is the only one that provides the surface enhancement effect with a single layer (Figure 2(a)). Not only is the signal intensity higher but more peaks and, therefore, more chemical information is present. On depositing multiple layers of 100 nm dilute colloid there is a change in the features present (Figure 2(b)). The intensity is increased and peaks become more apparent. However, after spraying 25 successive layers the improvement in quality was not sufficient to warrant the time involved. A similar effect and similar spectra were obtained for 100 nm colloid that had been pre-concentrated by a factor of 20 (Figure 2 (c)), therefore, pre-concentration also was not viable.

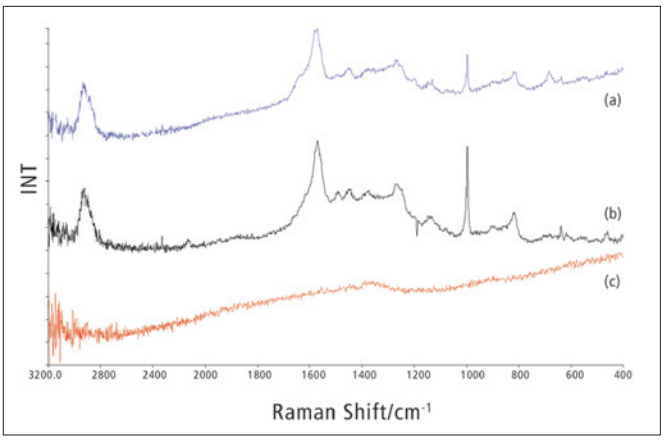


*Figure 3.* Spectra of (a) 100 nm dilute colloid, $Na_2SO_4$ and fingermark, (b) 100 nm dilute colloid and $Na_2SO_4$ only and (c) 100 nm dilute colloid and fingermark only.

### Salt Aggregates

Salt aggregates have been used with good results in other studies to improve the SERS enhancement effect.[3] Electrolyte salts are recommended since they do not bind strongly to the colloidal metal surface. Here sodium sulphate was used with the dilute 100 nm colloid to continue the investigation into limiting costs. Salt solutions at 0.1 M were mixed with the dilute 100 nm colloid in a 1:1 ratio. Figure 3 shows the spectra obtained for fingermarks with salt aggregates. As can be seen, the spectra of fingermarks with colloid and salt aggregates did result in an enhancement of some bands, however, by comparing these to spectra from the salt aggregate alone, the majority could be found in the salt aggregate spectra rather than being an improvement in the contribution from the fingermark. Additionally, peaks in the salt aggregate spectra overlap those in the fingermark spectra, thereby causing interference in peak assignment for fingermarks. Other salts were used and the same interference was observed (results not shown). Clearly, this interference is undesirable, particularly given the already complex nature of fingermark deposits. Therefore, the use of salt aggregation for signal enhancement was discounted, although other aggregating agents may be considered in the future.

### Order of deposition

Since multiple layers, pre-concentration and salt aggregates proved not to be beneficial for dilute colloids, attention was directed, for the remainder of the study, to the Raman specification 150 nm colloid. As previously discussed, any forensic method is ideally non-destructive. It is important, therefore, that a SERS enhancement is not gained at the expense of other techniques. Deposition of colloid is usually considered non-destructive, however, it could potentially interfere with subsequent enhancement. Spectra were taken of fingermarks where colloid had been deposited both before and after fingermarks (Figure 4). The spectra are shown on the same intensity scale and clearly show that higher intensity is achieved if colloid is deposited before the

fingermark. Fingermarks where colloid has been deposited before and after the fingermark were also enhanced with black powder and lifted. Both resulted in pattern detail suitable for comparison, however, where colloid had been deposited before the fingermark, ridges overlapping with the colloid were clearer. Subsequent analysis was carried out with colloid deposited before fingermarks.

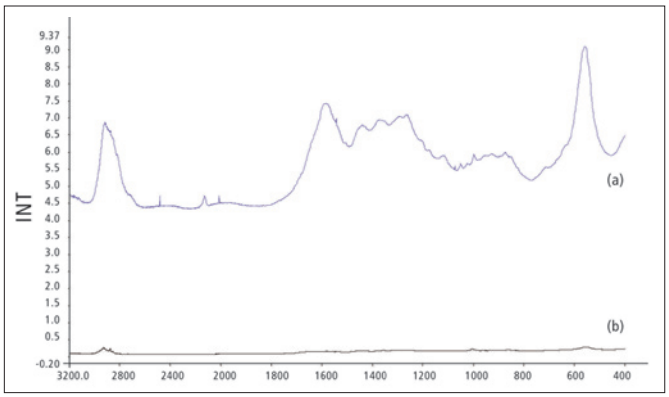


*Figure 4.* Effect of deposition sequence on fingermark spectra using 150 nm Raman specification gold colloid. (a) colloid deposited before fingermark and (b) fingermark deposited before colloid.

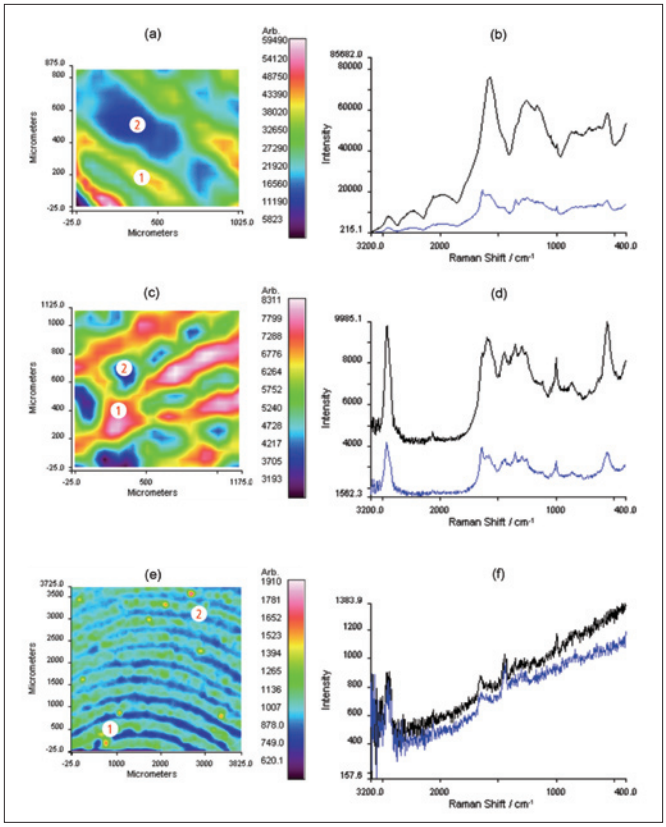


*Figure 5.* Fingermark images and corresponding spectra. (a) image on MALDI insert with 150 nm colloid, (b) spectra from positions 1 (black) and 2 (blue) on image 5 (a), (c) image on aluminium plate with 150 nm colloid, (d) spectra from positions 1 (black) and 2 (blue) on image 5 (c), (e) image on Klarite™ SERS substrate and (f) spectra from positions 1 (black) and 2 (blue) on image 5 (e). Images were obtained by integrating between 1010 and 990 $cm^{-1}$.

### Deposition surface

Images were acquired of fingermarks on three deposition surfaces, MALDI inserts, aluminium plates and Klarite™ pre-prepared SERS active substrate (Figure 5). For the MALDI insert and aluminium plate, the colloid was deposited by pipette resulting in a spot approximately 1.5 mm in diameter which limits the size of image that can presently be obtained. For the Klarite™ substrate, the active area was 4 mm x 4 mm allowing a larger, but still not full size, image. Image size will be addressed in future studies. As can be seen from Figure 5, each deposition surface contributes to the spectra. However, there are also differences between the deposition surface spectra and the fingermark spectra. Previous work on fingermarks and compounds present in fingermarks have assigned functional groups to peaks in the range examined here.[4] Day and co-workers[2] assigned peaks in "fingerprint oil" to C-H stretching and deformation modes (2899, 2851, 2728, 1439 and 1304 $cm^{-1}$) and to a C=C stretching mode (1661 $cm^{-1}$). In the current study, at around 1440 $cm^{-1}$ an unresolved, shoulder peak can be seen in the spectra of fingermarks on MALDI inserts and resolved peaks can be seen in the spectra from the fingermarks on aluminium plate and Klarite™. Day et al.'s work was carried out with a 633 nm HeNe laser without SERS enhancement, as opposed to the 785 nm laser used here, this would account for slight shifts in peak wavenumber. A peak at 1000 $cm^{-1}$ has been assigned by Virkler and Lednev[4] to the symmetric C-N stretch due to the presence of urea in sweat. For consistency, all images shown in Figure 5 were obtained by integrating scattering in the range 1010-990 $cm^{-1}$. In all images the distribution of C-N, from urea and other C-N containing constituents, allows the fingermark ridge pattern to be seen. Further work is required to remove deposition surface interference and assign peaks belonging to endogenous fingermark components.

## Conclusions

The technique presented here potentially provides a non-destructive method for fingermark analysis. Ridges have been successfully imaged on three surfaces, MALDI inserts, aluminium plate and Klarite™. Raman spectroscopy is already a portable technique, therefore, there is potential for combining it with a SERS substrate to provide immediate analysis at the scene. Ongoing work by the authors is focused on developing a cost effective method for retrieving full size fingermarks onto a SERS active substrate as well as investigating the detection of exogenous/contaminant components with this method.

## Acknowledgments

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