Analysing gunshot residues with Raman microscopy

Gunshot residues (GSR) are debris expelled from a discharged firearm. They often form a vital part of forensic examinations into firearm assaults. GSR are a complex mixture of particles derived from the propellant (burned and unburned inorganic and organic particles), primer components, and metals from the projectile or gun barrel[1].

Traditional GSR analysis identifies lead and other inorganic components, often by elemental analysis within a scanning electron microscope. However, this analysis has become more difficult with the introduction of lead-free ammunition. Forensic scientists therefore need new analytical methods that can study both organic and inorganic gunshot residues (OGSR and IGSR)[2].

The advantages of Raman spectroscopy

Raman spectroscopy is an excellent tool for studying GSR as it can analyse the chemical forms of both inorganic and organic GSR compounds, and to distinguish them from other environmental residues. Raman spectroscopy has other advantages: it is an optical technique that requires little sample preparation; and it is non-destructive, so evidence is preserved.

GSR are challenging samples. They consist of small hard-to-spot particles often on a substrate that gives its own chemical signal. The Raman system therefore needs a high-magnification research-grade microscope with the full range of optical viewing techniques, such as bright field, dark field, and polarised light. It also needs at least two lasers of different wavelength (ideally one in the visible spectrum and one in the infrared). Some particles will be fluorescent in certain parts of the spectrum and difficult to analyse on a single-wavelength system.

In the following example we illustrate the application of Raman spectroscopy to the analysis of gunshot residues. We used a Renishaw inVia™ Qontor® confocal Raman microscope, equipped with 532 nm (visible) and 785 nm (infra-red) lasers, and all the key optical viewing techniques.

Figure 1. Dark field optical image of GSR particles. The image is of a region 10.6 mm × 10.0 mm.

Preparing gunshot residues for analysis

The GSR were taken from a fired 9 mm calibre lead-free ammunition cartridge. We used a toothpick to gently dislodge traces from the interior of the cartridge onto a stainless-steel slide (these are ideal for Raman spectroscopy as, unlike standard glass slides, they do not give a significant Raman signal).

We viewed the particles with a 20× magnification objective lens. They ranged in size from about 100 µm to about 1 µm across. The larger particles could be seen using bright-field illumination, but dark-field gave better contrast for the small particles (Figure 1). The inVia Raman microscope was equipped with a Renishaw MS30 high-speed encoded microscope stage; this ensured precision and repeatability, essential when looking at small particles.

Analyse particles with ease

We used Renishaw's dedicated Particle Analysis (PA) software to automatically analyse the particles. This determines the particles' positions, sizes, and shapes from the microscope images, and then guides Raman data acquisition from them. You can then use the position and morphology values to restrict analysis to just a subset of the particles. The final result is detailed particle morphology and chemical information, obtained from just one simple experiment.

The PA software has a wide range of options for analysis. It can use a lower magnification lens to obtain the microscope images of the sample, then switch to a higher numerical aperture lens for more efficient, and therefore faster, Raman analysis. The following sections detail just one of the many analysis scenarios possible.

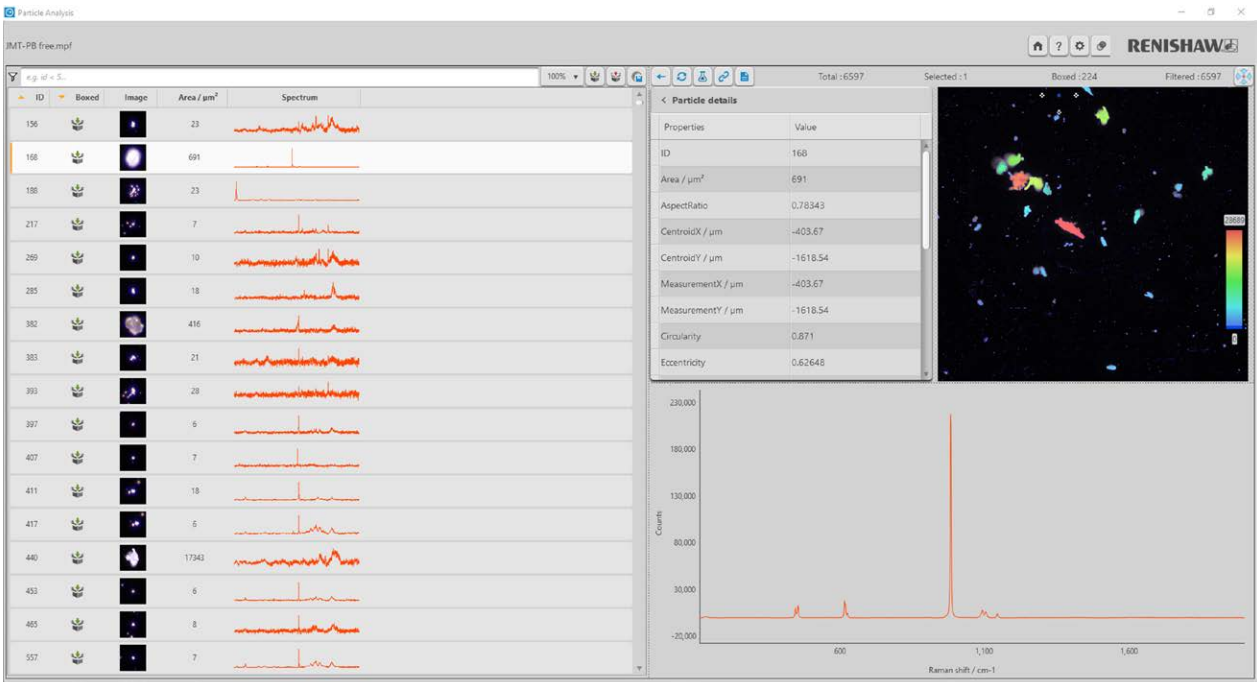


Figure 2. Analysing the GSR particles in the Particle Analysis software.

One simple workflow

We configured the PA software to survey the sample by optical microscopy, using dark-field illumination with a 20× magnification objective lens, and a 50× lens for the Raman measurements. A 532 nm laser excitation was primarily used for Raman analysis. This provided high quality data from the wide range of chemical compounds. Particles exhibiting high levels of fluorescence were reanalysed using 785 nm laser excitation.

The inVia Raman microscope was configured to use LiveTrack™ technology to automatically maintain focus on the particles. This enabled us to use a high-efficiency, high-magnification lens to rapidly acquire the Raman data. Without LiveTrack technology we would have had to use a lower-magnification lens with a bigger depth of field. This would have resulted in much lower optical efficiency and longer data acquisition times.

The PA software identified 6597 particles. The smallest of these were all features on the substrate, so we excluded any with areas below 6 µm². This gave 224 GSR candidates. The data from some of these is shown in Figure 2. Each analysed particle is listed with an ID number, optical image, particle area, and Raman spectrum. Highlighting the selected particle in the main table shows its detailed Raman spectrum, its position on the dark field image, and its morphology characteristics.

The PA software processes and analyses the Raman data automatically. In this case it removed cosmic ray artefacts and subtracted backgrounds. The chemical composition of the particles was determined by matching the processed spectra to reference data in Renishaw spectral libraries (other commercial libraries can also be used, if required).

Inorganic gunshot residues

This analysis confirmed the presence of different inorganic gunshot residue (IGSR) particles in the sample, including nitrates, carbonates, sulphates, pyrophosphates and oxides (Table 1).

Table 1. Inorganic particles (IGSR) confirmed by Raman analysis from lead-free ammunition.

	Nitrate	Carbonate	Sulphate	Pyrophosphate	Oxide
Strontium	Strontium nitrate	Strontium carbonate	-	-	-
Potassium	Potassium nitrate	Potassium carbonate	Potassium sulphate	-	-
Sodium	Sodium nitrate	-	-	-	-
Calcium	-	Calcium carbonate	-	-	-
Magnesium, calcium, iron	-	Calcium magnesium carbonate	-	-	Magnesium iron silicate
Magnesium	-	-	-	Magnesium pyrophosphate	-
Titanium	-	-	-	-	Anatase rutile
Zinc	Zinc nitrate	-	-	-	Zinc oxide

Most of the IGSR particles were present in the sample as pure inorganic compounds (Figure 3). The most abundant compound was calcium carbonate, which was found as a pure compound as well as a part of the mixture formed by different organic compounds in the organic gunshot residue (OGSR) particles. Furthermore, some of the GSR particles contained copper with isoindole-based organic compounds.

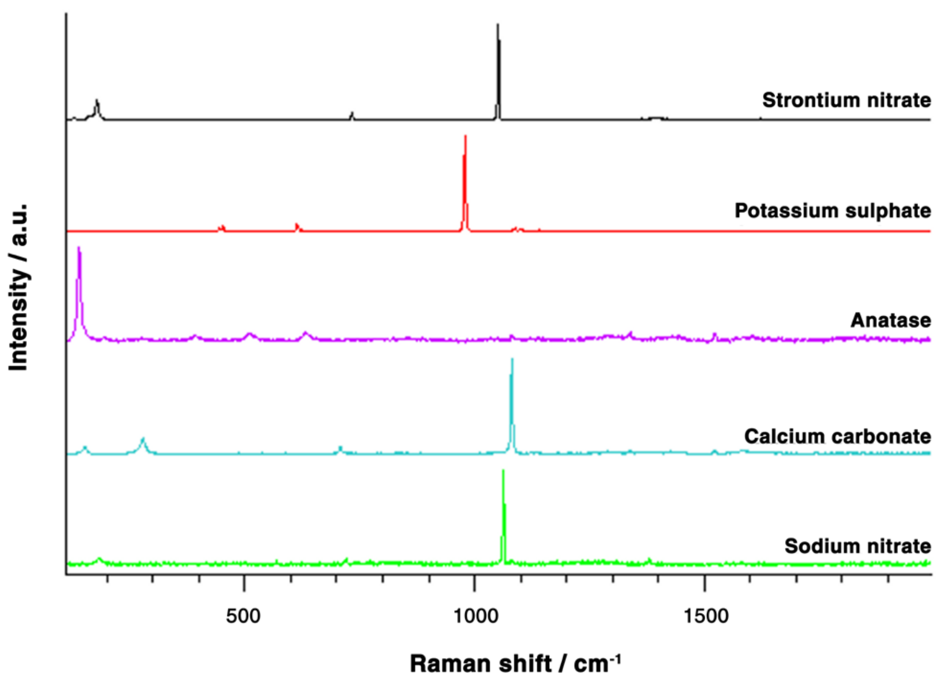


Figure 3. Example of IGSR Raman spectra collected from the fired cartridge.

Organic gunshot residues

The Raman analysis of organic gunshot residue (OGSR) particles is inherently more complex. This is because they are derived from organic compounds in not only propellants, but also weapon lubricants and by-products. They are also burned and transformed during firing[1].

Some of the organic compounds and functional groups detected in the gunshot residue were starch, quinoline, cresol, carbazole, naphthalene, anthracene, pirate, picric acid, styrene, nitro indazole, nitrobenzamide, dinitrobenzyl, diaminotrinitrobenzene, dinitrotoluene, graphite and other forms of carbon[3]. Compounds with azo- groups and sulphonyl- groups were also present.

Most of the OGSR Raman spectra were complex mixtures of different components. Figure 4 shows examples of spectra containing pure compounds, such as starch, and mixtures of IGSR and OGSR, such as calcium carbonate (bands at 153 cm^{-1}, 280 cm^{-1}, 711 cm^{-1} and 1085 cm^{-1}) and organic functional groups. Raman bands at 1346 cm^{-1} and 1590 cm^{-1} are characteristic for 2-nitrodiphenylamine and N-nitrosodiphenylamine, respectively. Both of those compounds correspond to smokeless gunpowder[4].

The Raman bands located at 1355 cm^{-1} and 1529 cm^{-1} correspond to NO_2 symmetric stretching mode and to NO_2 asymmetric stretching mode in nitrotoluene derivatives[5].

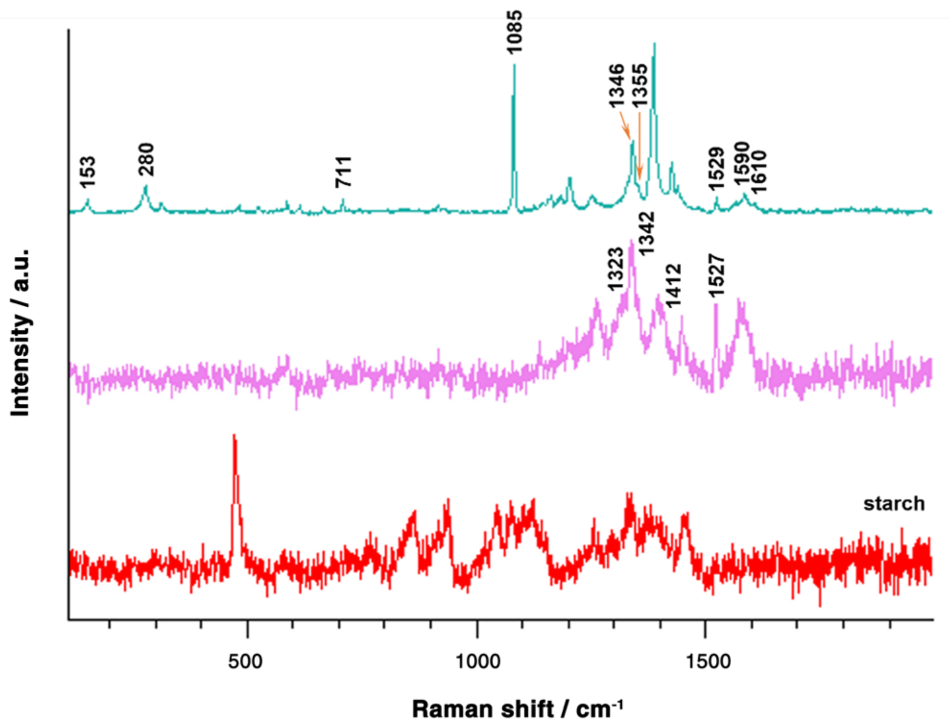


Figure 4. Examples of OGSR Raman spectra collected from the fired cartridge.

Flexible analysis

About 50 of the larger GSR particles exhibited fluorescence when excited with the 532 nm wavelength laser. We used the PA software to automatically collect Raman spectra of just these particles using the near-infrared 785 nm laser. The software presents these data alongside those from all the other particles analysed, in one combined dataset, making it easy to review the results.

The 785 nm Raman analysis showed the larger particles consisted mainly of cellulose, trans-ferulic acid or dimethoxystyrene.

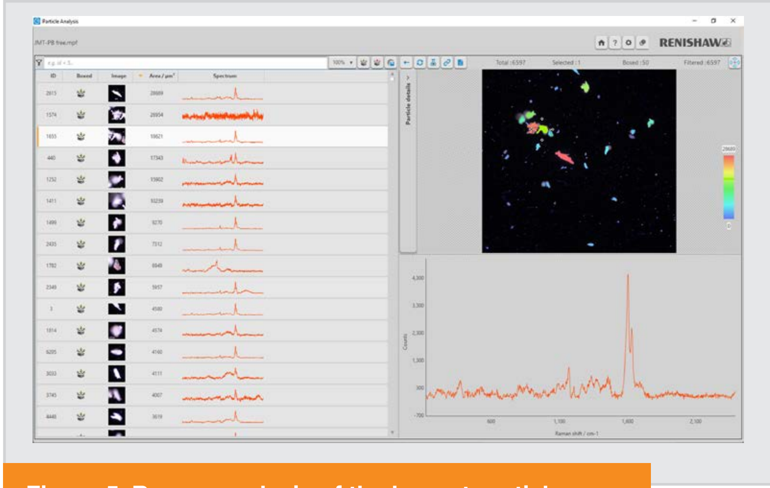
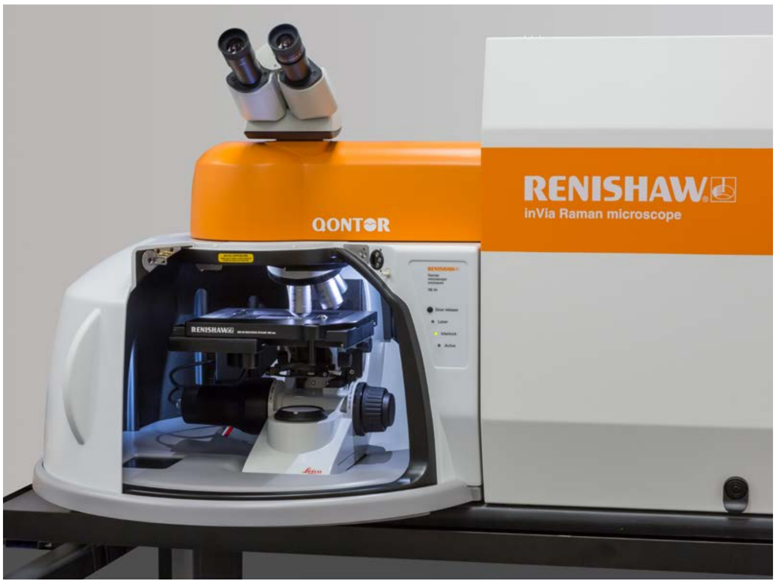


Figure 5. Raman analysis of the largest particles; data collected using 785 nm laser excitation.

The ideal tool for GSR analysis

Raman spectroscopy can deliver comprehensive morphological and chemical information about inorganic and organic GSR particles from a single, simple experiment. The research-grade inVia confocal Raman microscope, supported by the sophisticated PA software, provided extensive information on GSRs from the lead-free ammunition. This combination makes it quick and easy to obtain full information about inorganic and organic gunshot residues.



For more information on forensic applications of Raman spectroscopy, please visit **www.renishaw.com/forensics**

References

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