Noninvasive Authentication of Pharmaceutical Products through Packaging Using Spatially Offset Raman Spectroscopy

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We demonstrate the use of spatially offset Raman spectroscopy (SORS) in the identification of counterfeit pharmaceutical tablets and capsules through different types of packaging. The technique offers a substantially higher sensitivity than that available from conventional backscattering Raman spectroscopy. The approach is particularly beneficial in situations where the conventional Raman backscattering method is hampered or fails because of excessive surface Raman or fluorescence signals emanating from the packaging, capsule shell, or tablet coating contaminating the much weaker subsurface Raman signals of the active pharmaceutical ingredients and excipients held in the product. It is demonstrated that such interfering signals can be effectively suppressed by SORS.

There is an increasing need for the noninvasive verification of the authenticity of pharmaceutical products on the market. The number of generic copies of popular and well-known drugs is steadily increasing worldwide. The targeted drugs in the developed world often include so-called lifestyle drugs and drugs for chronic diseases. More seriously, in Africa and Asia, frequently the life-saving medicines, such as anti-infective drugs, are plagued by this problem. For example, the infiltration of the market by fake antimalarial drugs currently presents a major crisis in eastern Asia. Although the generic copy often has the correct molecule as its active pharmaceutical ingredient (API), the formulation of the drug can be very different, affecting the effectiveness of treatment. The worst case and potentially life-threatening scenario is when the generic product does not contain the supposed active ingredient at all. In recent years, large amounts of counterfeit drugs have been discovered and withdrawn from the official supply chain both in the UK and the United States, and the spread of drug sales over the Internet further exacerbates the problem. Terrorists are also believed to be partially funding their activities through the sale of counterfeit drugs over the Internet.3

Another alternative method is Raman spectroscopy, which exhibits exceptionally high chemical specificity. The method has also been used widely in the analysis of pharmaceutical products.5–12 Given the extent of the problem and its current trend, it is becoming increasingly important to verify the actual content of drugs throughout the entire supply chain. This task is complicated by the fact that once tablet packaging is opened, it becomes of no use, something which calls for a noninvasive method of analysis. Ideally, the method should be fast and provide clear and easy-to-interpret results. A handheld instrument is also highly desirable as many of these operations need to be performed in the field. One potential candidate technique for this analysis is near-infrared spectroscopy (NIR), which is a noninvasive method capable of providing results from whole tablets and capsules. Work has been done to use NIR for the noninvasive quality control of film coated and uncoated tablets.4 However, the subtle spectral differences in, for example, concentration between tablets, are often hard to detect and chemometric tools may be required in conjunction with this technique to provide clearer results. For this method to be effective, a prior knowledge of all potential subcomponents may be required. However, this may not always be possible with counterfeit drugs. The problem is also compounded by the fact that a relatively large variation in drug composition can exist between different production sites within the same company. In addition, NIR cannot always be deployed noninvasively because of the interference or absorption of NIR light by packaging material. Overall, NIR is considered to be a method better suited for a laboratory environment focusing on process and quality control rather than for the detection of counterfeit drugs in small local pharmacies and out in the field.

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spectra exhibit much sharper features compared with the NIR technique and provide accurate fingerprints of the analyzed molecules. The method in its conventional form permits the interrogation of many pharmaceutical products through their coating or capsules as well as through blister packs, and the technique has also been used to detect counterfeit drugs. However, in many instances, and in particular with dark-colored APIs, the Raman signal of API can be heavily polluted with fluorescence and Raman signals originating from the coating, capsule, or blister pack itself. These extra signals increase noise levels within the observed Raman spectrum of API and in some cases even preclude the observation of API and other ingredients held within the product altogether.

Recently, through collaborative efforts, we have developed a new form of Raman spectroscopy applicable to probing deep layers of turbid media, well beyond the reach of conventional Raman techniques such as confocal Raman microscopy. The method termed spatially offset Raman spectroscopy (SORS) provides qualitatively a new capability to analyze diffuse scattering media such as pharmaceutical drugs through their blister packs with much higher clarity than possible with conventional approaches. In parallel research, it has been demonstrated that transmission Raman spectroscopy can also be used very effectively for non-invasive probing of the bulk content of pharmaceutical products.

The SORS approach is based on the collection of Raman spectra from spatially offset regions away from the point of illumination on the sample surface and subsequent scaled subtraction of the spectra (or multivariate data analysis) to separate the signals of individual layers within the interrogated sample. Since the first demonstration of the SORS concept, the technique has been used in numerous applications including Raman tomography in turbid media by Morris et al., the noninvasive Raman spectroscopy of bones on cadavers and animal samples by Morris et al., and the first observation of human bone in vivo under safe illumination conditions by our collaborative team.

This work is performed in the basic SORS point illumination and collection geometry. The method was chosen for its simplicity and compatibility with differently shaped pharmaceutical products and also because there are no severe intensity restrictions on the illumination intensities with pharmaceutical products necessitating otherwise the use of more elaborate geometries such as inverse SORS. It should be noted that the abovementioned SORS variants, as well as the transmission geometry, however, could also be deployed in many of these applications. The use of transmission geometry may not be straightforward with blister packs and plastic jars, since the sample may not be easily accessible from two opposing sides, as is required with this technique, or the sample may be too thick. For example, blister packs are often coated with a metallic film on one side and are therefore not transparent to the laser beam and Raman signal. In contrast, the one-side metal coating does not pose any problem for a SORS geometry that requires access from one side of the packaging only. This permits not only the probing of blister packs but also diffusely scattering plastic containers. The type of packaging used may vary geographically from region to region, often just for cultural reasons. For example, blister pack is the preferred and most sold packaging type for common drugs in the UK, but in the United States, it is far more common to sell these drugs in plastic jars. For this reason, we demonstrate the performance of the technique on both types of packaging.

MATERIALS AND METHODS

Samples. Common over the counter coated tablets and capsules containing predominantly paracetamol or ibuprofen were purchased. Care was taken to ensure that a wide variety of tablet coatings and capsule colors was represented. Nurofen caplets (Crooke’s Healthcare) were small white ibuprofen-containing tablets and Solpadeine Max (GlaxoSmithKline) had a red coating and a white interior with paracetamol and codeine phosphate hemihydrate as active ingredients. The capsules purchased were Sudafed Dual Relief and Sudafed Congestion Relief (yellow/green and yellow capsules, respectively, Pfizer) and Lloyd’s Pharmacy Ibuprofen (blue/clear capsule, Healthy Ideas). The active ingredients were paracetamol, caffeine, and phenylephrine hydrochloride for Sudafed Dual Relief; phenylephrine hydrochloride for Sudafed Congestion Relief; and ibuprofen only for Lloyd’s Pharmacy capsules. Plain paracetamol tablets (Christy Products, M&A Pharmachem Ltd.) packed in a plastic jar were also purchased.

Apparatus. The Raman spectra were measured using a SORS Raman apparatus described in detail earlier. The probe beam was generated using an attenuated 115-mW, temperature-stabilized diode laser for Raman spectroscopy, operating at 827 nm (Micro Laser Systems, Inc, L4 830S-115-TE). The laser power at the sample was 50 mW, and the laser spot diameter at the sample was ~0.5–1 mm. The beam was spectrally purified by removing any residual amplified spontaneous emission components from its spectrum using two 830-nm bandpass filters (Semrock), and the beam was incident on the sample at ~45°. The spatial offset used throughout the SORS experiments was ~3 mm. Controlled conventional backscattering Raman measurements were performed using the same instrumentation with spatial offset set to zero.

Raman light was collected in backscattering geometry using a 50-mm-diameter collection lens with a focal length of 60 mm. The scattered light was collimated and passed through a 50-mm-diameter holographic notch filter (830 nm, Kaiser Optical Systems, Inc.) to suppress the elastically scattered component of light. The second lens, with the same parameters as the collection lens, was then used to image, with magnification 1:1, the sample interaction zone onto the front face of the fiber probe. The Raman light was collected in the basic SORS point illumination and collection geometry.
propagated through the SORS annular fiber systems of length ~2 m to the linear fiber end oriented vertically and placed in the input image plane of a Kaiser Optical Technologies Holospec f = 1.8i NIR spectrograph with 167-μm-wide slit. Raman spectra were collected using a NIR back-illuminated, deep-depletion, TE-cooled CCD camera (Andor Technology, DU420A-BR-DD, 1024 × 256 pixels) by binning the entire chip vertically to produce a single Raman spectrum for each illumination condition of a given spectral offset. The Raman spectra shown are not corrected for the variation of the detection system sensitivity across the spectral range.

Unlike in our previous work, 10 here we used a fiber bundle-probe made of 22 tightly packed active fibers at the center of the probe. The signal from these was binned into a single Raman spectrum in each measurement for a given spatial offset as indicated above. The spatial offset was introduced by altering the position of the incident laser beam on the sample as illustrated in Figure 1. The individual fibers were made of silica with a core diameter of 220 μm, a doped silica cladding diameter of 240 μm, and a polyimide coating of 265-μm diameter. The fiber numerical aperture was 0.37. The bundle was custom-made by CeramOptec Industries, Inc.

Raman Measurements. For the noninvasive SORS measurements, the unperturbed blister packs were taken out of their cardboard packaging and placed on the sample stage of the Raman instrument so that the illuminating laser light impacted on the rounded part of the blister pack containing the capsule or coated tablet. An identical sample arrangement was used for the conventional Raman measurements. The resulting spectra were then compared with the reference spectra obtained by breaking the coated tablets in half and placed in such a way that the illuminating laser light was incident on the surface of tablet interior. This provided an accurate reference Raman signal accounting for the presence of both the active ingredients and excipients held within the tablet, enabling the comparison of noninvasive Raman spectra with those obtained by invasive means. In a similar manner, the capsules were opened and the contents transferred to a fused-silica cuvette and analyzed. Tablets with an indent or imprint were measured on the side with no indent or imprint. Acquisition time was 1 s, and 10 separate accumulations were taken and averaged for each spectrum. The emptied blister packs were also analyzed in order to identify any spectral contribution originating from the packaging.

RESULTS AND DISCUSSION

Blister Pack Packaging. All tablets and capsules were packed in white semitransparent blister packs in which it was just about possible to see the outline of any brightly colored content. Common excipients were talc, magnesium stearate, starch, and TiO₂, with TiO₂ giving rise to intense interfering Raman signals. The thickness of the capsulés was on average 0.16 mm. The conventional Raman and SORS setup geometries are schematically represented in Figure 1A, and a photograph depicting the analyzed drugs and their respective packaging is shown in Figure 1B.

Figure 2 shows the SORS and conventional Raman spectra of four different tablets and capsules; Sudafed Dual Relief capsules, Solpadeine Max tablets, Nurofen caplets, and Lloyd’s Ibuprofen capsules. The green and yellow capsules of Sudafed Dual Relief gave rise to a fluorescent background signal in both the conventional and SORS spectrum (Figure 2A). The conventional Raman spectrum (CR) has many spectral features that correspond well with that of the reference spectrum, but there are also several intense spectral bands that can be assigned to the blister pack. The blister pack features are indicated with asterisks (*): these overlap with several important spectral bands of the API. The SORS spectrum has an overall lower signal intensity and thus a poorer signal-to-noise ratio, but nevertheless, there is a very good correspondence with the tablet reference (ref) as Raman signals originating from the packaging are very effectively suppressed. The triplet API band at 611, 631, and 656 cm⁻¹ is detected in the SORS spectrum whereas this feature is unresolved and polluted with spectral contribution from the blister pack itself in the CR spectrum. The API band at 700 cm⁻¹ is also overlapped and broadened by blister pack signal so that it cannot be used for identification in the CR spectrum. Other similar overlaps between blister pack and API are also present at 1435 (blister pack) and 1449 (API) cm⁻¹ and at 458 (blister pack) and 471 (API) cm⁻¹.

The Raman spectrum of Solpadeine Max tablet is also dominated by paracetamol, which is apparent through the almost identical reference spectrum (Figure 2B). The CR spectrum has the same unresolved and overlapping problems as Sudafed Dual Relief, precluding definite identification of the material within capsule. Again, the SORS spectrum does not suffer from either of these overlaps as the packaging signal is effectively suppressed. The SORS signal quality is also higher, yielding a very good match, which permits positive identification of API between the SORS and API reference spectra.

Nurofen caplets (Figure 2C) and Lloyd’s Ibuprofen capsules (Figure 2D) both contain ibuprofen as the only active ingredient. Ibuprofen is a weaker Raman scatterer compared to paracetamol, which might explain the lower quality of Raman spectra. However, a positive identification of ibuprofen was clearly possible with the SORS spectrum. This is in sharp contrast with the corresponding CR spectrum, which suffers from spectral interference with the Raman signal of the blister pack. The spectral region 900–1400 cm⁻¹ of the CR spectrum shows major overlaps between ibuprofen and blister pack. This area remains poorly unresolved and contributes to the difficulty of identifying the API using the CR spectrum of Nurofen. In all, the CR spectrum has few isolated ibuprofen spectral bands; in general, they are partially or completely overlapped by blister pack signal. This is especially apparent at the prominent ibuprofen doublet at 1180 and 1206 cm⁻¹, which is heavily polluted with blister pack signal. For the SORS spectrum on the other hand, there is a perfect match between the reference and SORS spectrum. Two very weak minor
blister pack overlaps could be identified at 650 and 1450 cm$^{-1}$, but these do not alter the overall pattern of the spectrum and do not hamper a correct identification of the API.

Both the conventional Raman and SORS spectrum of Lloyd’s ibuprofen capsules have significant fluorescent background signals that make it more difficult to detect the ibuprofen spectral bands (Figure 2D). The ibuprofen triplet close to 1100 cm$^{-1}$ and doublet at 1200 cm$^{-1}$ are both present in the SORS spectrum, though weak, making the spectral band pattern of the SORS spectrum identical to the reference. The CR spectrum shows a similar pattern although there are also some overlaps between blister pack and ibuprofen spectra contribution at lower wavenumber.

Another benefit of SORS is its low sensitivity toward the movements of the tablet or capsule inside the blister pack. Conventional Raman measurements are likely to produce results that vary due to these movements giving rise to variation in relative spectral intensities between the blister pack and the pharmaceutical product, which in turn could be wrongly misinterpreted as variations due to counterfeit origin.

Sudafed Congestion Relief capsules, with phenylephrine hydrochloride as API, provided a major fluorescence background signal for both the conventional and spatially offset Raman spectra (Figure 3A). The main spectral contribution of the CR raw spectrum was from the blister pack itself, with a few weak Raman bands from API. The spectral bands of the SORS raw spectrum were all relatively weak (as phenylephrine hydrochloride is a weaker Raman scatterer compared with paracetamol), but the match with the reference spectrum is clear. A straightforward point-to-point background subtraction removed the fluorescent background signal enabling easier identification of API (Figure 3B). For the CR spectrum, the background subtraction enhanced the spectral bands originating from the API but also those originating from blister pack, yielding some overlaps. The overall spectral pattern of the CR spectrum does not resemble that of phenylephrine hydrochloride. On the other hand, the SORS spectrum, although rather noisy, exhibits spectral features that match well those of the reference spectrum. However, some contribution originating from the blister pack still remains around 720 cm$^{-1}$. A point-to-point background spline subtraction was performed in Grams/AI software (Thermo Galactic) although it should be noted this procedure is subjective and can introduce distortions to the spectra. In practical application, we would envisage the use of either a polynomial curve-fitting method or an automated routine.\(^\text{23}\)

**Plastic Container Packaging.** In order to further demonstrate the potential of using SORS for the detection and identification of pharmaceuticals through their packaging, the detection of medicines packed in thicker plastic bottles and jars was also demonstrated. Plain paracetamol tablets packed in a standard white plastic jar of a wall thickness of 1.7 mm with a snap-off lid were analyzed directly through their packaging. Figure 4A shows the conventional and spatially offset Raman spectra together with the tablet reference and that of the jar itself. Both sample spectra contain paracetamol component and varying degrees of spectral contribution originating from the jar. The reference and jar spectra have many bands adjacent to each other that overlap in both the conventional and SORS spectra. Some of these overlaps are


**Figure 2.** Conventional Raman and SORS spectra of four different coated tablets and capsules: (A) Sudafed Dual Relief capsules, (B) Solpadeine Max tablets, (C) Nurofen caplets, and (D) Lloyd’s Ibuprofen capsules. Major and other significant spectral bands of the active pharmaceutical ingredient are marked with dashed lines and spectral contributions originating from the corresponding blister packs are indicated with asterisks. Legend: CR, conventional backscattering Raman spectrum; SORS, spatially offset Raman spectrum; ref, reference spectrum.
indicated by dashed lines in Figure 4A. The Raman band at 517 cm$^{-1}$ of the tablet reference lies close to a slightly stronger band at 543 cm$^{-1}$ in the jar spectrum. The conventional Raman spectrum has a single spectral band at 543 cm$^{-1}$ whereas the SORS spectrum has a doublet that matches the positions of both the reference and the jar. The major triplet feature of the paracetamol spectrum at 800, 835, and 860 cm$^{-1}$ is present in both the CR and SORS spectra. However, the jar spectrum has a doublet at 810 and 844 cm$^{-1}$, which affects the sample spectra. The CR spectrum is more similar to the jar spectrum as the small 835-cm$^{-1}$ paracetamol band is not present at all. The SORS spectrum has the same appearance as the paracetamol spectrum but also shows clear overlaps with the features originating from the jar spectrum. At 1152 cm$^{-1}$, the jar spectrum has a broad feature that overlaps with the 1169-cm$^{-1}$ band of the reference. Again, the CR spectrum is dominated by the jar reference and the SORS spectrum by paracetamol. Band broadening also occurs for both CR and SORS spectra at 1369 cm$^{-1}$, and a single jar feature shows up in both spectra at 1458 cm$^{-1}$. The latter shows that despite the spatial offset there still is a significant contribution to the SORS spectrum originating from the jar. Nevertheless, the overall spectral pattern of the SORS spectrum is dominated by paracetamol whereas the conventional Raman spectrum resembles that of the jar.

To take the full advantage of the SORS concept, a scaled background subtraction was made. The zero offset spectrum was scaled using a scaling factor of 0.103 and subtracted from the 3-mm offset spectrum to cancel the surface Raman signal contributions, thus enabling the complete rejection of any spectral contributions originating from the plastic jar (Figure 4B). This step is not possible with a conventional Raman approach, which cannot provide two such Raman spectra with different relative intensities between the surface and subsurface signals. As can be seen, the relative peak intensities, ratios between peaks, and the overall spectral pattern of the corrected SORS spectrum match extremely well that of the API reference spectrum. The doublet at 517 and 543 cm$^{-1}$ is now a paracetamol singlet at 517 cm$^{-1}$ (indicated by the dashed line). The triplet at 800–860 cm$^{-1}$ now also better reproduces the reference spectrum as the jar contribution is completely removed.
CONCLUSIONS
The work demonstrates the improved sensitivity of spatially offset Raman spectroscopy over conventional backscattering Raman spectroscopy in the identity testing of pharmaceutical products through packaging. The new approach is particularly beneficial in situations where the conventional Raman backscattering method is hampered or fails because of excessive surface Raman or fluorescence signals emanating from the packaging, capsule shell, tablet coating, or plastic container that contaminates the much weaker subsurface Raman signals of the active pharmaceutical ingredients and excipients held in the product with undesired noise. These interfering signals can be effectively suppressed by SORS.

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