P-SERS™ — Trace detection overcoming the cost and usability limitations of traditional SERS technology

Introduction

Approaches to detect minute amounts of molecules have historically been expensive and time-consuming. Recently, surface enhanced Raman spectroscopy (SERS) has emerged as a practical trace detection technology enabled by a portable detector and a cleverly-engineered substrate.

The need for highly sensitive detection is pervasive across a wide range of industries, including:

- Food contamination
    - Melamine in milk
    - Unsafe pesticide levels
- Anti-counterfeiting
    - Weakened/inactive counterfeit drugs
    - Counterfeit perfumes and liquors
- Drug or explosive residue detection (e.g. at ports of entry)
    - Cocaine, heroin, THC

Each of these challenges, and many more, can be addressed by trace detection technologies, yet in practice the high costs and complexity can limit their use and effectiveness.

SERS is faster and easier to perform compared to the most commonly used techniques such as those which use mass spectrometry and gas chromatography, so why isn't it more widespread? The leading challenge in the contemporary use of SERS is that substrates tend to be either expensive ($10's-$100) or have sensitivity and repeatability issues.

Here we provide a brief description of SERS technology and offer a new, alternative approach to SERS substrates which maintains measurement sensitivity and reliability while solving issues of cost and complexity. A number of application examples are highlighted to assist in evaluating these claims.

Why SERS?

When molecules are excited by light, some photons are Raman scattered. Raman spectroscopy measures the unique fingerprint-like spectrum of this scattered light to identify unknown molecules. While Raman spectroscopy has long been performed to analyze bulk materials, it has very poor sensitivity and is easily overwhelmed by sample fluorescence. SERS

> *"**SERS identifies molecules by their fingerprint-like spectra, enabling detection of targets into the parts per trillion in complex samples**"*

intensifies the Raman emission through the use of a plasmonic enhancing material, boosting the signal by millions of times and enabling detection limits as low as parts per trillion levels.

Benefits of SERS

While the current widespread trace detection technologies offer similar detection performance, SERS has a number of appealing features:

- Inexpensive, portable equipment
- Simultaneous detection of multiple sample constituents
- Minimal false positives
- Portable, detection at the point of sample collection

P-SERS brings additional benefits, which together create a compelling argument for the use of SERS:
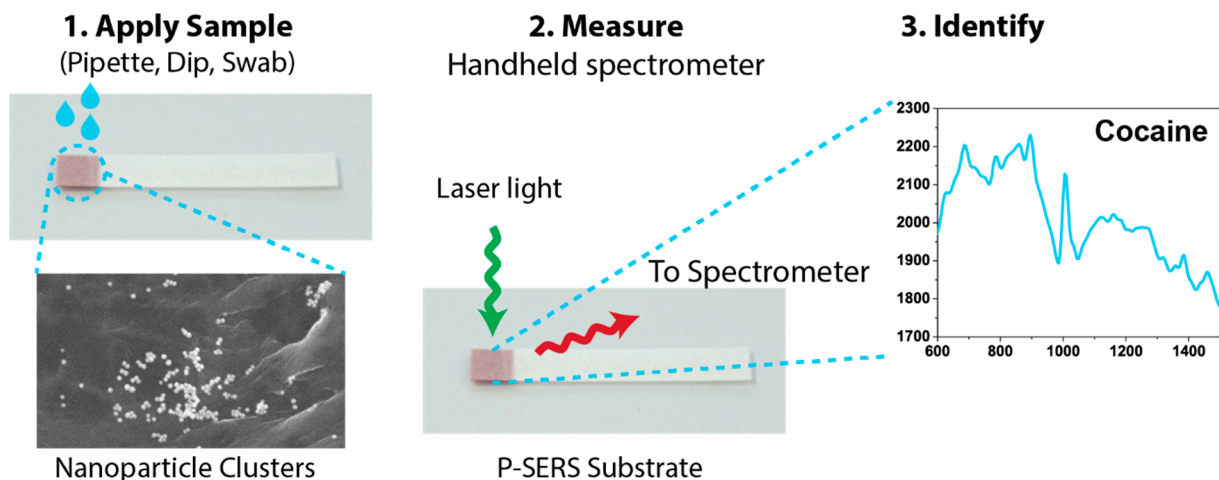
- Low per-sample costs
- Detection in seconds
- Mitigates fluorescence issues found in real-world samples
- Sample collection and cleanup is an intrinsic capability of the substrate itself

Substrates enable sensitive, reliable detection

Performing SERS measurements requires two components: a portable spectrometer and a plasmonically active surface: *a substrate*. The substrate provides the signal enhancement and provides inter-sample consistency. While portable spectrometers are readily available and increasingly affordable, most substrates are produced using clean-room lithography and are prohibitively expensive. P-SERS offers a way to generate substrates in a reliable yet relatively inexpensive manner.

SERS measurements using a substrate

Although traditional substrates are expensive and require manual sample application and drying, P-SERS enables flexible and quick sample application in the form of pipetting or a simple dip or swab. Following sample application, measurements can typically be performed without drying, yielding results in under a minute. To measure the substrate a portable spectrometer is used. The sample is then identified by its spectrum, either manually or automatically by the spectrometer software.

1. Apply Sample
(Pipette, Dip, Swab)

Nanoparticle Clusters

2. Measure
Handheld spectrometer

Laser light

To Spectrometer

P-SERS Substrate

3. Identify

Cocaine

P-SERS substrates: a low-cost, high enhancement solution

Unlike commercial cleanroom microfabricated substrates, P-SERS substrates are formed using a patent-pending process to deposit nanostructures through ink-jet printing, lowering the cost dramatically while maintaining uniformity and enhancement.

An additional advantage of the ink-jet process is that P-SERS substrates can be formed on a variety of membrane materials, not just silicon or glass wafers. Cellulose paper is typically used, but the material can

"Ink-jet fabrication for low-cost, flexible and high enhancement SERS substrates"

be altered to suit a particular application. Generating substrates in this manner opens up a number of new ways to use and optimize these substrates:

1. Pipetting the sample onto the planar surface (as with "traditional" substrates)
2. Dipping to absorb the sample into the substrate
3. Swabbing a surface to collect a trace residue
4. Changing the substrate material itself for one-step cleanup and detection (e.g. using membranes which bind interferents instead of cellulose paper)
5. Using lateral flow to separate a sample from interferents

Each of these use cases will be explored in depth in this section, with relevant application examples.

P-SERS as a traditional substrate

While P-SERS substrates have a number of unique properties which will be explored in later application examples, they are also high-performance substrates in their own right, both in terms of sensitivity and measurement reproducibility.

A substrate can be used on its own as a paper strip or mounted to a glass slide. To take a measurement, a sample is simply placed onto the substrate and then a spectrometer is used to measure the SERS spectrum. A wide range of chemicals and potential contaminants can be detected, from explosives and narcotics to pesticides and dyes. Likewise, tags or tracers added to detect counterfeiting can be easily detected.
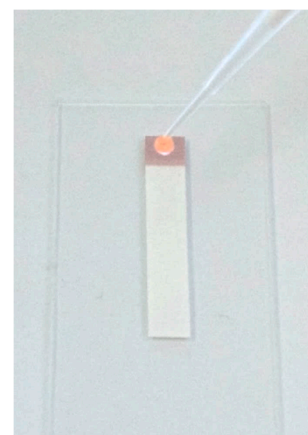
P-SERS substrates offer a number of properties which we believe make them especially suitable for use in routine analysis:

- Immediate measurement – no need to dry for most analytes (as with many SERS substrates)
- Substrates can be generated in any size and shape, on a variety of materials
- P-SERS substrates show high measurement reproducibility
- Internal standards can be incorporated for further improved quantitative accuracy (inquire for details)

Application Example 1a
Trace chemical marker detection for materials authentication and anti-counterfeiting

A common tracer molecule, BPE (1,2-Bis(4-pyridyl)ethylene), is added as a tracer to a liquid. Then, a droplet of the liquid (2 µL in this case) is added to the P-SERS substrate and measured. The concentration of the BPE molecule can be used as a tracer to ascertain the authenticity of a liquid. Substituted liquids would lack the tracer signal entirely, while liquids that have been "cut" would have a lowered signal intensity. We have shown reliable detection down to 1.8 ppb (~4 picograms of BPE in total), measuring an average 1207 cm^{-1} of 33 counts/s using a portable fiber optic spectrometer[1]. This concept could be applied to many products, from pharmaceuticals to cosmetics.
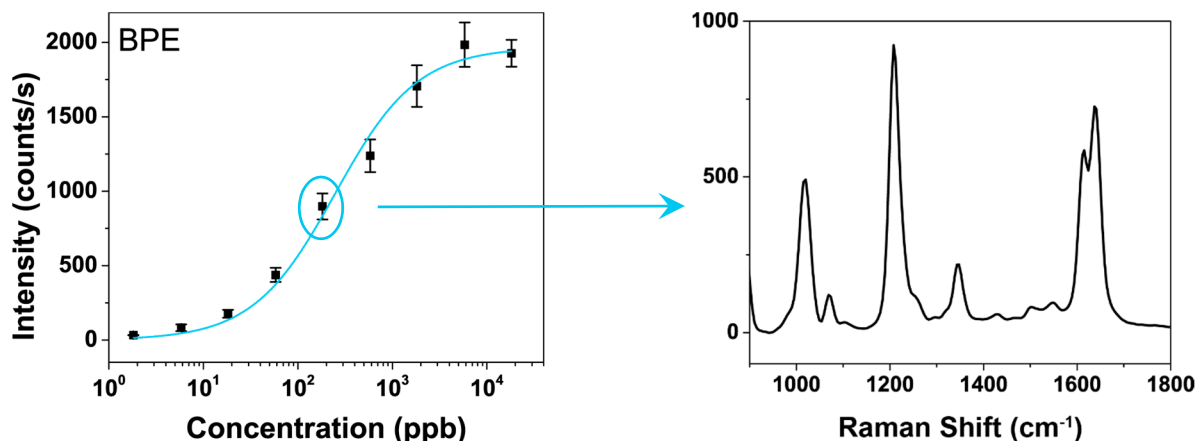

Technical details:
The plot on the right shows a measured SERS spectrum for 182 ppb BPE. For the plot on the left, the signal strength is quantified using the height of the 1207 cm^{-1} peak height. This was repeated six times for each conentration, and the error bars represent one standard deviation of the 1207 cm^{-1} peak height measurements. The curve fitted to the left plot is a langmuir isotherm, with an R^2 of .99, indicating a very high quality of fit. Nearly all analytes we have worked with can be fitted with the same curve to generate analyte-specific calibration curves.

Application Example 1b
Trace explosive detection

Here we present an example of explosive detection to further demonstrate the trace detection capabilities of P-SERS substrates.

This spectrum was obtained by applying a solution containing 1 microgram of TNT to a P-SERS substrate.
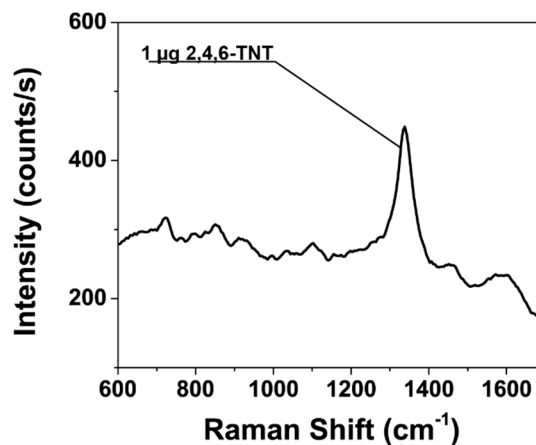
Sample loading through swabbing and dipping

Traditional substrates are rigid and must be manually loaded by applying a droplet with a pipette and waiting for it to dry. While P-SERS substrates can effectively serve this need, they are also much more versatile:

- Samples can be loaded by dipping
- Sample concentration is possible by extending the dipping time
- Trace residues can be collected by swabbing a surface
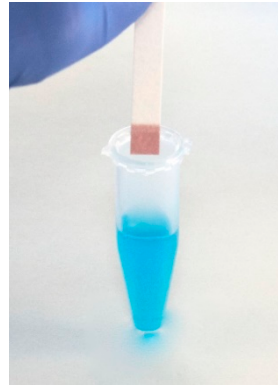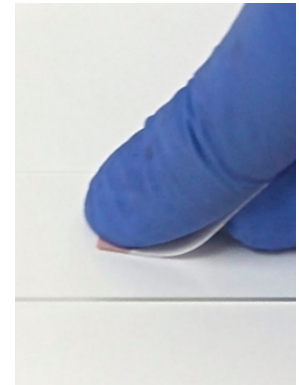
Dip   **Swab**
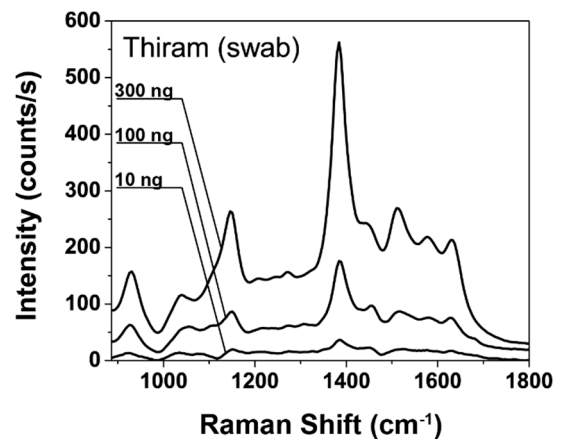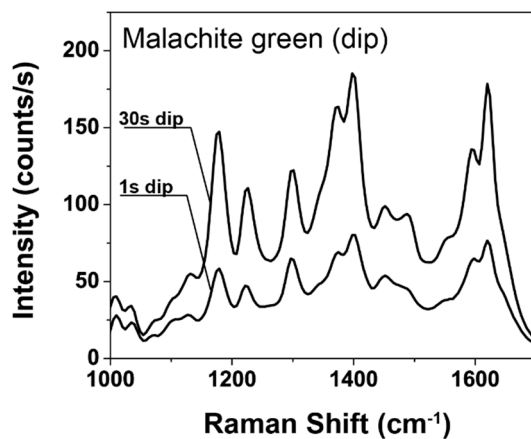
Application Example 2

Dipsticks and swabs for pesticide and fungicide detection

Due to the flexible nature of P-SERS substrates, they can double as both wipes for sample collection and as SERS sensors, enabling screening for a wide variety of substrates, from pesticides and pollutants to narcotics and explosives. Here we present the detection of two different fungicides, using P-SERS substrates in both dipstick and swab formats[1].

Malachite green is a banned fungicide, yet nevertheless is periodically detected in imported food, particular from fish farms. Here, we shows SERS spectra for a P-SERS dipstick dipped into a solution of water containing *1 ppb malachite green* (left figure). The signal is immediately measured without drying.

"…enabling screening for a wide variety of substrates, from pesticides and pollutants to narcotics and explosives"

The fungicide Thiram is often used to prevent crop damage (especially fruits). Here, we show the results obtained by swabbing a surface with a known amount of Thiram deposited on it. A clear signal can be seen even *as low as 10 nanograms* (right figure).

Application Example 3

Cocaine residue surface swab (e.g. from a table, bag, hand)

Here we present detection of 5 µg of cocaine residue on a smooth surface obtained by swabbing the surface with a P-SERS sensor. In past work, we have found that an easily measurable signal was present at the limit of detection of 25 nanograms (100 counts/s) [2]. Sensor performance is similar for detection of heroin residues.
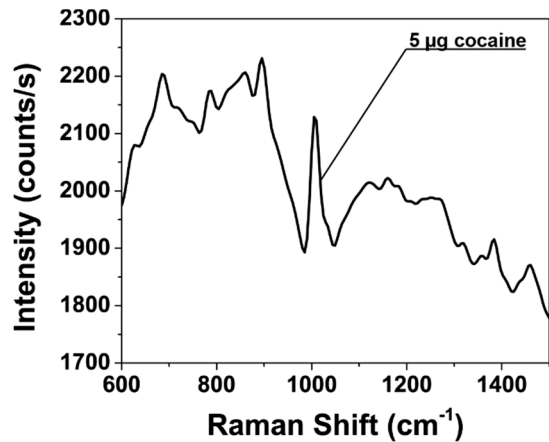
On-sensor sample cleanup

Since P-SERS substrates can be fabricated on a variety of membranes, they can also serve as separation and concentration matrices for detection of targets in difficult sample matrices. They also integrate well into lateral-flow style assays.
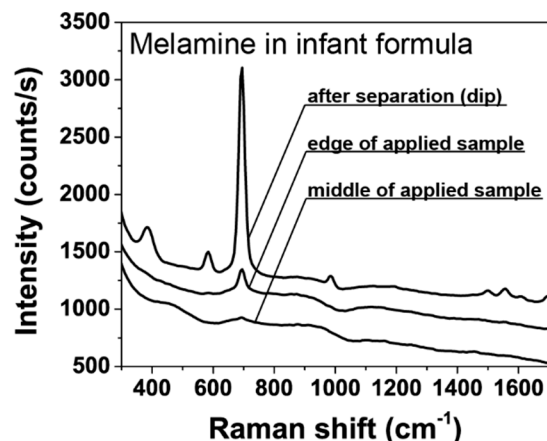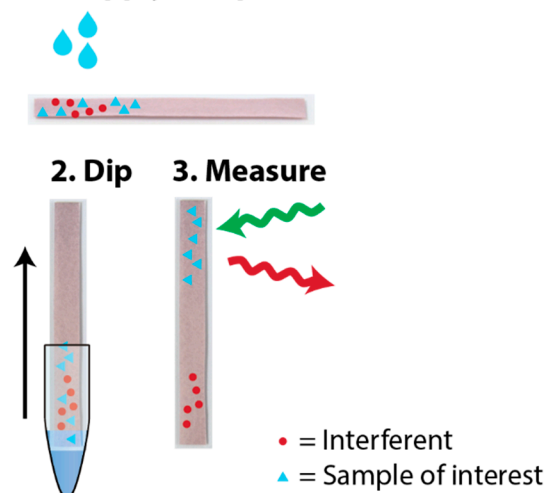
Application Example 4

On-substrate sample cleanup for melamine detection in infant formula

Food products present one example of complex sample matrices, so the ability to use the substrate to separate chemical targets from the bulk materials in the food (proteins etc) is particularly useful. By choosing a protein binding membrane, the proteins which would otherwise foul the sensor surface are bound in place and the analytes of interest remain free to flow to the printed detection region.

"Interestingly, the inherent wicking has already begun to separate the melamine from the interferents on its own…"

Here we show results obtained by using PVDF P-SERS substrates for detection of 100 ppm melamine in milk[3]. The PVDF material binds the protein interferents and allows for easy detection of the melamine. The bottom trace is obtained at the location of sample application. The middle trace is collected at the edge of the applied sample: interestingly, the inherent wicking has already begun to separate the melamine from the interferents, enabling clear detection of the melamine signature. A dip provides complete separation (top signal).

1. Apply Sample

2. Dip **3. Measure**

• = Interferent
▲ = Sample of interest

Application Example 5
Separation and measurement of narcotics mixed with fluorescent cutting agents

While Raman can be used for identification of pure illicit drugs, oftentimes drugs are cut with agents that fluoresce, preventing detection of these street narcotics. The inherent ability of P-SERS substrates to separate the sample through chromatography overcomes this problem, enabling detection of the target analyte in an otherwise fluorescent sample. The principle is the same as the separation depicted in Application Example 4.

Here we present before and after spectra for a sample of heroin which was spiked with a fluorescent dye[3]. Initially, the signal is entirely overwhelmed by the fluorescence. However, after a dipping into a methanol-acetic acid solution (in which the narcotic is soluble and thus mobile, while the fluorescent cutting agent is not), a clear signal for the heroin can be detected.
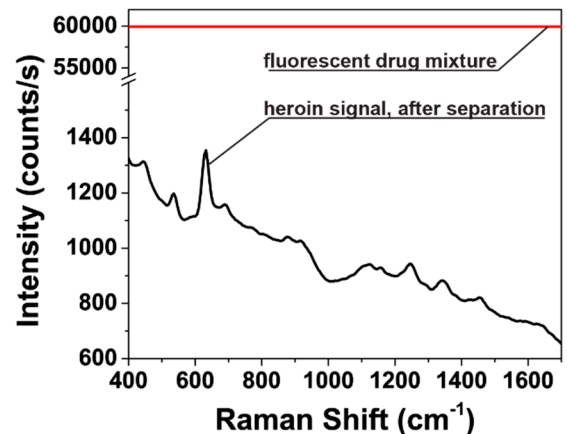
Conclusion
P-SERS offers the benefits of SERS without many of the assay format and use limitations of traditional rigid substrates.

SERS is a natural fit for many trace detection applications, from detection of narcotic, pesticide and explosive residues to applications including anti-counterfeiting, R&D and industrial process control.

While SERS has historically been prohibitively expensive, P-SERS dramatically lowers costs to dollars per sample while also enabling entirely new test formats, including swabs and dipsticks: formats which are simply not possible with traditional substrates!

Make P-SERS work for you
Don't just take our word for it:

> *Visit us on the web at www.diagnosticanSERS.com to get your introductory kit*

We have extensive experience optimizing SERS assays for a variety of applications. We also work with partner organizations to custom tailor SERS readers to your needs, whether those needs require R&D bench-top style setups or a simple instrument for "red light / green light" field use.

Have a particular application that requires a detection unique approach?

> *Contact us to discuss custom engineering of P-SERS substrates,*
> *and how P-SERS can solve your trace detection needs*

We look forward to hearing from you.

How to contact us

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References

[1]   E. P. Hoppmann, W. W. Yu, and I. M. White, "Highly sensitive and flexible inkjet printed SERS sensors on paper," *Methods*, vol. 63, no. 3, pp. 219–224, 2013.

[2]   W. W. Yu and I. M. White, "Inkjet-printed paper-based SERS dipsticks and swabs for trace chemical detection.," *Analyst*, vol. 138, no. 4, pp. 1020–5, Feb. 2013.

[3]   W. W. Yu and I. M. White, "Chromatographic separation and detection of target analytes from complex samples using inkjet printed SERS substrates.," *Analyst*, vol. 138, no. 13, pp. 3679–86, Jul. 2013.