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3D SERS Platforms: Large-scale Plasmonic Hotspots for New Applications in Sensing, Microreaction and Data Storage.

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1. INTRODUCTION

Surface-enhanced Raman scattering (SERS) is a molecular spectroscopy method that utilizes plasmonic nanostructures, mainly of Ag and Au, to significantly enhance the normally weak Raman vibrational spectrum of molecules.[1,2] In general, SERS provides up to 10^{10}-fold Raman enhancement via two main mechanisms; electromagnetic enhancement and chemical enhancement.[3] The former is the major contributor for SERS enhancement and arises from the collective oscillation of the metal's conduction band electrons upon interaction with light.[4] This generates intense electromagnetic (EM) field at metallic surfaces that amplifies the scattered Raman signals of target molecules up to 10^8-fold. Chemical enhancement accounts for up to 10^3 Raman signal enhancement that arises from the increased polarizability as a result of charge-transfer between the chemisorbed molecules and the plasmonic metal surface.[5] Importantly, SERS is able to bring about characteristic vibrational fingerprints of the target molecules and sensitivities down to the single molecule level, which makes it superior to other analytical techniques such as fluorescence and chromatography.[6] Hence, SERS has been applied as a reliable technique for quantitative detection, reactivity investigation and reaction mechanisms elucidation at the molecular-level.[7]

Over the last four decades, a great deal of efforts has been devoted to the design and fabrication of new types and configurations of SERS platforms in order to adapt to various sensing scenarios from fixed static measurements to dynamic sensing in fluids (either liquids or gases).[8] While some individual nanoparticles (NPs, zero-dimensional (0D)) may provide highly localized hotspots at single-particle level for fundamental studies, experimental evidences suggest that the best SERS enhancements arise from nanoparticle clusters owing to interparticle plasmonic coupling.[9-11] While these platforms allow detection at single molecule level, they suffer from large SERS signal fluctuations within a SERS substrate, thus inhibiting quantitative applications.[12] Consequently, over the last 15 years, there have been increasing emphasis on using 2D plasmonic platforms comprising of organized nanoparticle assembly to improve SERS' signal sensitivity and reproducibility.[13] These platforms have been developed into many substrate-based variations, fabricated by a wide range of techniques including colloidal imprint lithography,[14,15] self-assembly,[16] or Langmuir-Blodgett.[17]

Recently, 3D SERS platforms with additional plasmonic material in the z-axis are fabricated with the aim to achieve 3D hotspots for even higher SERS enhancements. These 3D platforms bring about several advantages over the conventional 2D planar platforms. Firstly, the larger hotspot volume takes advantage of the inherent 3D space of the Raman excitation laser focal volume, which have depths ranging from sub-micrometers to several centimeters.[18] The 3D SERS hotspots within the laser volume are larger than their 2D counterparts, which significantly increases SERS signals.[19] Moreover, increasing the surface area of 3D platforms can further enhance the interactions between target molecules and plasmonic particles hence improving detection sensitivities. 3D SERS platforms also have an advantage in terms of practicality. Its higher hotspot density in z-axis allows better tolerance to laser misfocus, providing more feasible and timely on-site measurements without tedious laser alignment prior to detection.[20] In contrast to conventional 2D platforms, 3D platforms give the flexibility to construct plasmonic materials in all spatial directions with varying sizes and shapes.[21] This results in the design of various 3D structures for a series of SERS-based applications that extend beyond sensing purposes such as hierarchical plasmonic-active pillars for optical storage via z-dependent SERS imaging,[22] substrate-less capsules for microreactions and reaction monitoring,[23] and plasmonic cloud for in-air measurements.[24] The evolution from 2D to 3D in SERS platform design and fabrication broadens the horizons of SERS research, and transforms SERS from a microscope-based sensing method into a practical and multipurpose read-out technique that can play a major role in future technology.

In this account, we introduce our most recent works on unconventional 3D plasmonic platforms and focus on their unique sensing and non-sensing applications that arise from their 3D configurations (Table 1). Herein, we define 3D platforms as those having hotspot volumes ranging from sub-micrometer (≥ 0.5 μm) to the centimeter-scale, because they complement most Raman spectrometers with laser focal depths ≥ 0.5 μm.[25] We first highlight our library of contemporary 3D platforms and classify them into two main categories of substrate-based and substrate-less platforms (Figure 1). Subsequently, we showcase the unconventional applications of 3D SERS platforms that are superior to their 2D counterparts, in both sensing and beyond sensing applications. We aim to provide a scientific synopsis and insights on the current and future trends in the design and applications of 3D SERS platform, which bring promises to new scientific discoveries and technology developments in 3D SERS platforms for tackling real-world issues.

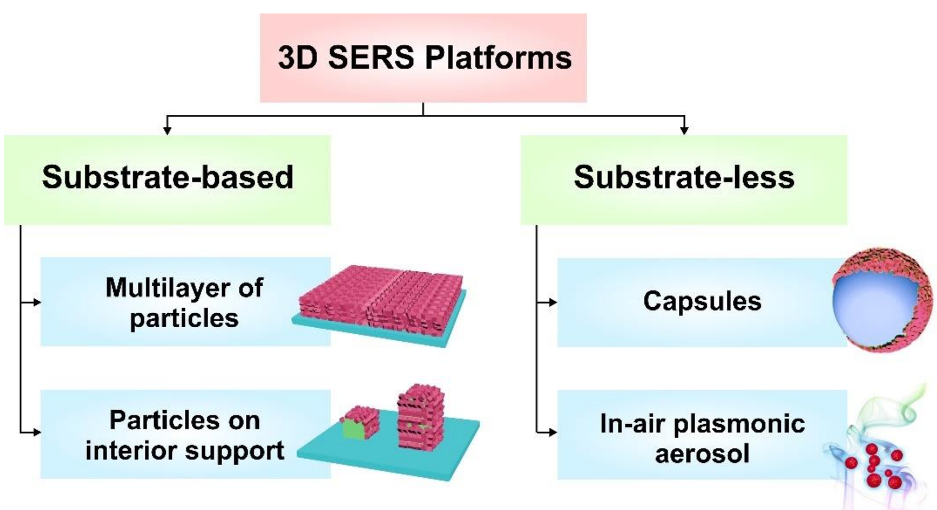

Figure 1. An overview of 3D substrate-based and substrate-less SERS platforms.

Table 1. Fabrication, hotspot dimension, sensing medium, enhancement factor and applications of various substrate-based and substrate-less contemporary 3D SERS platforms.

3D SERS platform ^ Plasmonic building block	Fabrication approach	Hotspot dimension in the z-axis	Enhancement factor (EF)/ Analytical enhancement factor (AEF)	Application	Ref
Substrate-based platforms					
Multilayer of particles					
Gold nanorod supercrystal ^ Au nanorod	Sendimentation	1 µm	10^{6} * EF	Sensing	27
Dual-structure assembly ^ Ag nanocube, octahedra, cubeoctahedra, truncated cube	Self-assembly in drying droplet (Bottom-up)	10 µm	5.2×10^{6} * EF	Sensing	29
Mesh-like structure ^ Ag nanowire	Langmuir-Schaefer (Top-down)	1 - 3.2 µm	1.8×10^{11} * AEF	Sensing	32
Particles on interior supports					
Plasmonic pyramidal and prism array ^ Ag nanocube	Lithography-writting of polymeric structures and coating of Ag	5 - 15 µm	2.6×10^{6} *EF	Data storage	33
Candlestick array ^ Ag film	Lithography-writting of polymeric structures and coating of Ag	1.75 - 6.75 µm	2.1×10^{6} *EF	Data storage	22
Substrate-less platforms					
Capsules					
Plasmonic liquid marble (PLM) ^ Ag nanocube	Self-assembly of superhydrophobic Ag particles	1 mm - 1 cm	5×10^{8} * AEF	Microreactions/ Multiphase sensing	35
Plasmonic colloidosomes ^ Au/Ag nanoparticle	Self-assembly of superhydrophobic Ag/Au particles	0.5 - 150 µm	10^{7} *AEF	Microreactions/ Multiphase sensing	36
In-air plasmonic aerosols					
Plasmonic aerosol ^ Ag nanocubes/ colloidosomes	Spraying suspension of plasmonic colloidosomes via nebulizer	up to 2.3 cm	1.8×10^{5} * AEF	Stand-off in-air sensing	24

2. SUBSTRATE-BASED 3D SERS PLATFORMS

Substrate-based SERS platforms refer to plasmonic nanostructures assembled/fabricated on a flat solid support – typically silicon wafer or glass slide. This support provides mechanical stability to the SERS platform, and enables better fabrication reproducibility, which is desirable for the mass production of SERS chips for analysis.

2.1 Multilayered particles on solid substrates

One of the most common methods to create 3D hotspots is to increase particle height in the z-direction by building multilayered plasmonic nanoparticles on a solid substrate. In contrast to nanoparticle aggregates, these supercrystals are highly ordered nanoparticles which provides a high and homogenous electromagnetic field.[26,27] The most straightforward way to produce supercrystals is via the sedimentation of a droplet of concentrated nanoparticles solution on a flat surface, which forms 3D supercrystals upon drying (Figure 2A).[28] Using this method, we obtain an extended multilayered nanorod supercrystal of ~1 µm of 3D hotspot thickness which yields high SERS enhancement factor (EF) of $\sim 10^6$.[27] Beyond conventional uniform supercrystal fabrication, we demonstrate the concept of one assembly with dual crystal structures using Ag polyhedral nanoparticles, achieved due to the collective effect of large particle edge size and surfactants (Figure 2B).[29] Two distinct crystalline structures with total thicknesses of 10 µm are achieved by creating two different self-assembled micro-environments within the droplet: one at the drying front and another at the air/water interface. This results in a more open structure with high orientational order at the air/water interface and a close-packed structure with high translational-order at the drying front within the droplet. Notably, the formation of dual-structure supercrystals results in a synergistic SERS enhancement as compared to the uniform supercrystal;

Ag octahedra dual-supercrystals achieve a SERS EF of 5.2×10^6 which is 3.3-fold higher than the uniform supercrystals.

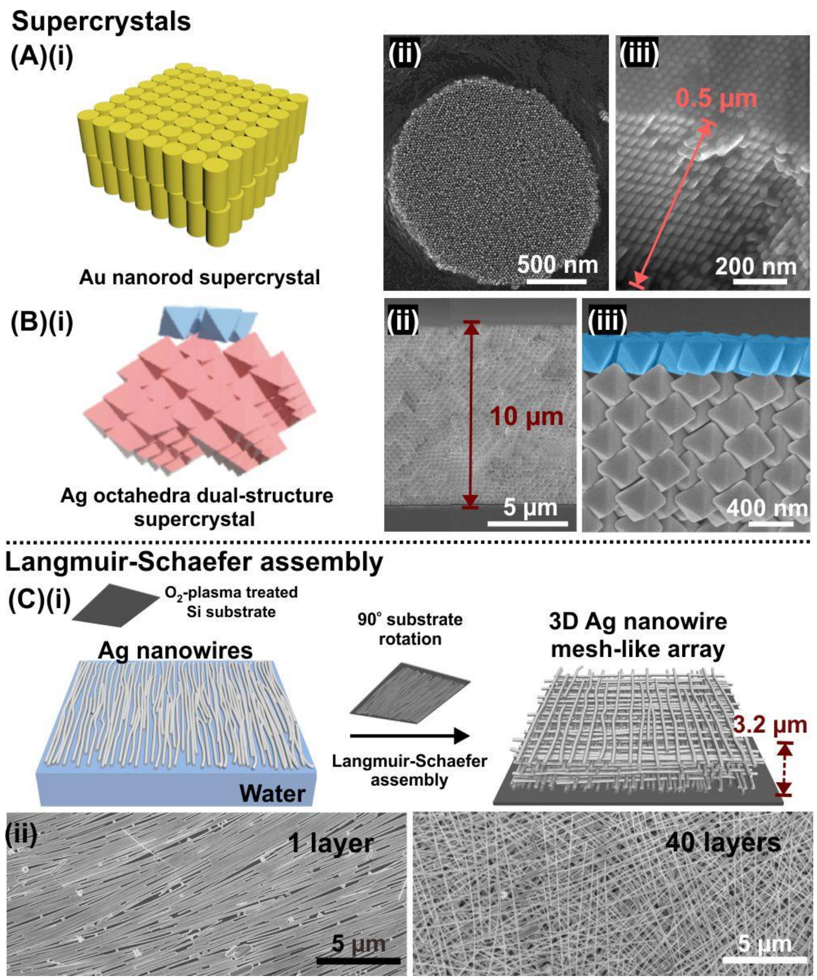

Figure 2. **Fabrication of multilayered 3D structures.** (A)(i) Scheme and (ii, iii) SEM images of Au nanorod supercrystal island film. Reproduced with permission from ref 27, Copyright 2011 National Academy of Sciences. (B)(i) Scheme and (ii, iii) SEM images of dual-structure Ag octahedra supercrystal. Reproduced from ref 29. Copyright 2018 Nature Publishing Group. (C)(i) Scheme of self-assembly of multilayered nanowire structures using Langmuir-Schaefer. (ii) SEM images of close-packed and increasingly mesh-like nanowire arrays at 1 and 40 layer. Adapted with permission from ref 32. Copyright 2014 American Chemical Society.

However, supercrystals present certain limitations. The quality of supercrystals relies extensively on the preparation of very homogeneous nanoparticles. These nanoparticles also need to be highly concentrated, which is prone to aggregation. Another challenge in droplet-based assembly is the need to precisely control the evaporation conditions, preferably without any perturbation over a long period of time (hours to days). Moreover, the close-packed nature of these multilayered substrates inhibits lasers to penetrate beyond ~2 μm substrate thickness.[30,31] Hence, the resulting EF plateaus after reaching the laser penetration threshold.

To overcome the laser penetration bottleneck, we fabricate open 3D multilayered structures that allows deeper laser penetration using Langmuir-Schaefer (LS) assembly technique. It involves the successive stacking of NP monolayers assembled at the air/water interface onto a substrate (Figure 2C).[32] For example, we fabricate up to 40 layers of open mesh-like structures of Ag nanowires (diameter of 85 nm) with an overall thickness of ~ 3.2 μm (Figure 2C). This platform achieves a high analytical enhancement factor (AEF) of up to 1.8×10^{11}, which is 8-fold higher than close-packed woodpile-like assembly of the same nanowires, demonstrating the ability of the laser to penetrate through the open structure.

Overall, multilayered NP assembly is one of the simplest methods to organize nanoparticles into micron-scale 3D structures and exploit the collective effect of 3D plasmonic clusters to improve the EF. A common drawback is the low particle usage efficiency, where only the outermost nanoparticle layers are accessible for interaction with the target analyte molecules.

2.2 Three-dimensional hotspots on interior supports

An efficient alternative to multilayered substrates that reduces wastage of plasmonic materials is by depositing plasmonic nanoparticles on non-plasmonic supporting templates. The well-defined supporting templates impart the 3D structural configuration, whereas the plasmonic particles provide the SERS properties. Such method is more economic because the supports are made from more abundant and cost-effective materials such as polymeric materials, which reduces the use of noble metals.

The interior supporting structures can be fabricated using lithography methods, e.g. nanoimprint lithography or laser photolithography.[33,34] As for the plasmonic coating, physical methods such as vapor deposition and sputtering, or colloidal methods such as layer-by-layer deposition or self-assembly can be employed. One highlight of our works features polymeric pyramidal structures coated with Ag film and Ag nanocube assembly, which achieve high hotspot dimension from 5 to 15 μm in height (Figure 3A-C).[33] These 3D pyramidal nanostructures exhibit hotspots in three orthogonal directions, giving rise to an EF of 2.6×10^6. Besides pyramids, the flexibility in designing supporting structures with lithography also allows us to construct 3D plasmonic prismatic arrays with various heights from up to 15 μm across the substrate, which is applicable as data encoding platform via z-dependent SERS imaging (Figure 3D).

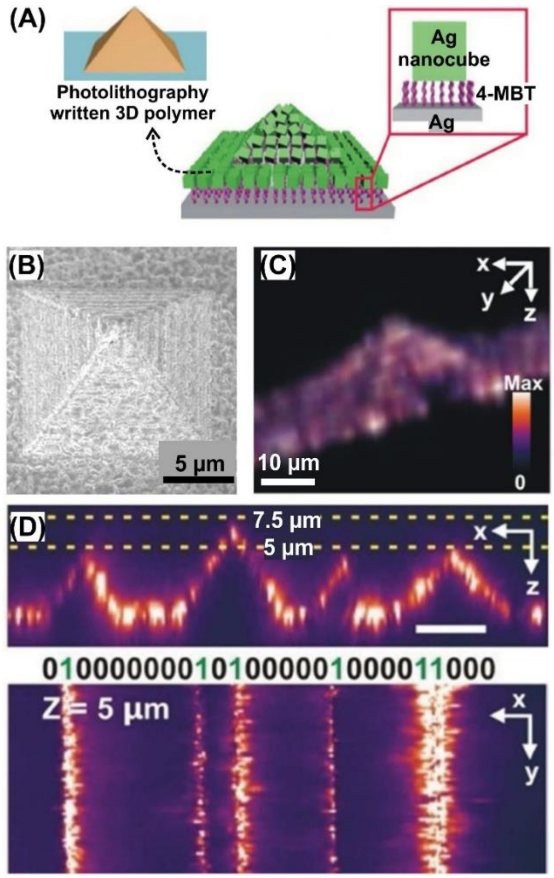

Figure 3. **3D hotspots on interior supports**. (A) Scheme of 3D pyramidal structures coated with Ag and Ag nanocubes. (B) SEM image and (C) 3D hyperspectral SERS image of plasmonic pyramid structures. (D) X-y and x-z hyperspectral SERS image of a plasmonic prism array. Adapted with permission from ref 33. Copyright 2014 John Wiley & Sons, Ltd.

In summary, this approach of depositing plasmonic materials on 3D interior supports has a more economical particle usage than constructing multilayered plasmonic structures, because only the outermost layer is made of plasmonic materials. Another advantage lies within the ability to create arbitrary 3D structures with controllable shapes and sizes via lithography technique, which is highly challenging in supercrystal formation or LS assembly. The trade-off, however, is the additional step needed to fabricate these supporting templates.

3. SUBSTRATE-LESS 3D SERS PLATFORMS

Substrate-less platforms are standalone structures not bound to any solid support. They are generally more mobile than their substrate-based counterparts, and thus can be incorporated within fluid/fluid interfaces, solution or even aerosols. Furthermore, these standalone platforms are accessible to analyte and excitation laser from all spatial directions, which is desirable for novel multiphasic applications. This section showcases our unique substrate-less 3D SERS platforms ranging from solution-based plasmonic microcapsules to air-based plasmonic aerosols with hotspot spanning over centimeter scale.

3.1 Substrate-less plasmonic capsules

One emerging substrate-less 3D SERS platform is plasmonic capsule, a soft platform whereby plasmonic particles are assembled at the air/liquid or liquid/liquid interface of a droplet in order to minimize the surface energy.[35] We are the first to fabricate plasmonic capsules, including millimeter-scale plasmonic liquid marbles (PLM) and subsequently micrometer-scale plasmonic colloidosomes with picolitre volumes (Figure 4). Both platforms are formed by encapsulating aqueous droplets with Ag nanoparticles, and have > 10 layers of close-packed Ag particles in their shells to give rise to homogeneous and highly SERS-active surfaces for ultrasensitive SERS sensing with an analytical enhancement factor (AEF) over 10^8.[35,36]

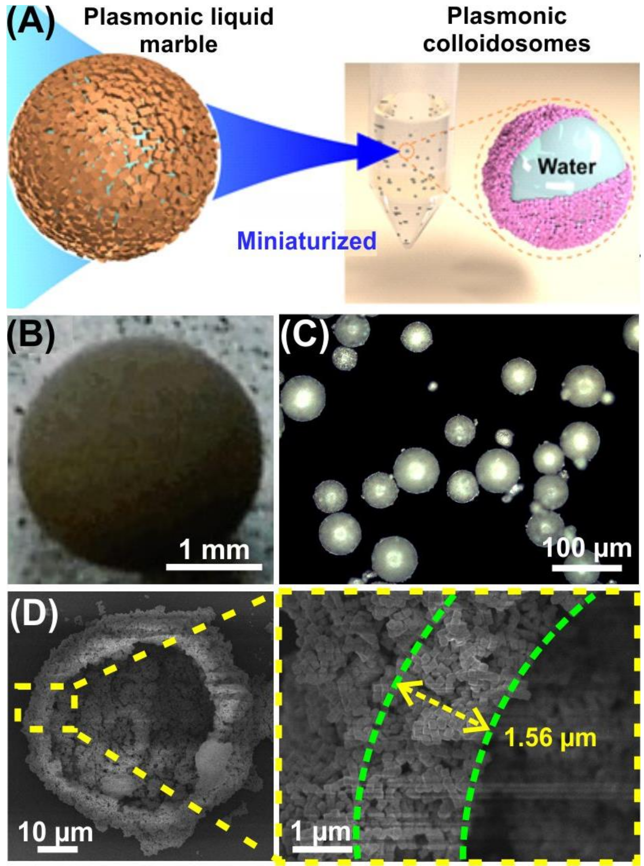

Figure 4. **Substrate-less plasmonic capsules** (A) Schemes of plasmonic liquid marble (PLM) and plasmonic colloidosomes. (B) Digital image of a PLM. Reproduced with permission from ref 35. Copyright 2015 John Wiley & Sons, Ltd. (C) Microscopic image of plasmonic colloidosomes. (D) SEM images of the self-assembled colloidosomes and the inset shows the corresponding high-magnification SEM image of the multilayered shell structure. Reproduced with permission from ref 36. Copyright 2016 John Wiley & Sons, Ltd.

Notably, the capsules can also be immersed in oil phase to achieve water-plasmonic nanoparticle-oil interface, and this system allows the multiplex detection of ultratrace analytes across the water-oil interface. We also demonstrate that the plasmonic colloidosome shells are permeable and enable *in situ* SERS observation of analyte diffusion across the interface that is challenging using other analytical techniques. Such interfacial property is unique to this class of substrate-less SERS platforms, and highly advantageous for the SERS study of interfacial phenomena at the molecular level. Collectively, the miniatured 3D spherical shape and permeable shells of these plasmonic capsules make them attractive for isolating and monitoring single- or dual-phase microreactions in a non-invasive manner (section 4.2).[37-40] While we have been fabricating water-in-oil capsules thus far, moving forward, we envision that the future development of other emulsion systems such as oil-in-water capsules can bring forth immense potential in multiphasic analysis, which are important for food, color and cosmetic applications.

3.2 In-air plasmonic aerosols

While solution-based detections have been widely developed, the detection of aerosolized/gaseous species directly in air remains extremely challenging due to their dynamic nature, random diffusion and low concentrations. In-air 3D aerosol SERS platform, a dispersion of plasmonic nanoparticles in the air, emerges as a viable solution because it allows direct interaction of the plasmonic materials with airborne species to give instantaneous SERS read-out. This averts the substantial waiting time needed for the analyte to accumulate onto the SERS-active surface when employing substrate-based platforms or capsules.

Recently, we disperse aerosolized plasmonic colloidosomes (APCs) for direct SERS measurements of airborne analyte in their native in-air environment (Figure 5).[24] This colloidosome-infused aerosol cloud brings about an effective plasmonic hotspot of ~2.3 cm in length, the largest yet achieved, to allow detection within a large volume. Our platform maximizes direct interactions between the plasmonic nanoparticles with the gaseous analyte, thus greatly increasing the SERS detection signals.[24] Additionally, the plasmonic aerosol's large centimeter-scale volume translates to very high tolerance to laser misfocus and achieves 100-fold higher tolerance to laser misalignment compared to conventional 2D substrates.[26] This unique feature is advantageous for gas or vapor detection because of their high mobility, and the large hotspot volume provides flexibility to adjust or maneuver the focal point right at the airborne species, eradicating the need for substrate alignment.[41] This also gives the plasmonic aerosols immense potential to couple with stand-off Raman devices for remote sensing, where precise laser focus is challenging. 3D plasmonic aerosols are also omnidirectional, which enable efficient detection of analyte introduced from any direction, independent of laser angle alignment. Notably, our system achieves EFs over 10^5 and obtains detection limit of methylene blue aerosols down to 0.5 nmol.

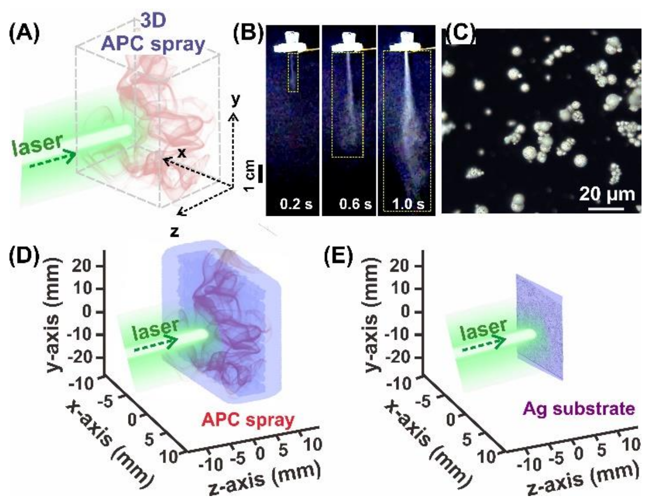

Figure 5. Aerosol-based 3D SERS platforms. (A) Illustration of the spread of APCs (with 4-MBT) in the x, y, and z directions. (B) Digital image of the colloidosome mist dispensed from an aerosolizer within a 1 second timeframe. (C) Microscopic image of aerosolized colloidosomes collected in a hexane solution after spray. (D-E) Comparison of SERS detection capabilities between (D) APCs and (E) 2D substrate based on different parameters; SERS-active volume along the x-, y-, and z-axes. Reproduced with permission from ref 24. Copyright 2018 John Wiley & Sons, Ltd.

While APC marks a key milestone to promote the progress of in-air SERS, improvements can be made by devising aerosols that can suspend in mid-air for longer interaction duration with the gas species to further improve the sensitivity and reproducibility of the system. Moving forward, we also foresee conferring selectivity to the plasmonic aerosols to be the key to avert strong background interferences. Such additional properties will significantly improve the overall sensitivity and signal stability during in-air SERS signal acquisition of a highly dispersed and dynamic airborne sample.

4. EMERGING APPLICATIONS OF 3D SERS PLATFORM

4.1 Chemical sensing

The sensing of chemicals that reveals information about their molecular composition, concentration, and chemical activity is crucial in multiple fields such as medicine, human health, food safety, homeland security and environment. Our 3D SERS platforms with special design and structural diversity exhibit prominent SERS performance, thus have been utilized in chemical sensing across a wide range of environments from static to flowing in liquid, or even in dynamic airborne conditions.

4.1.1 Close range sensing

Close range SERS sensing relies on confocal microscope objectives to acquire signals within millimeter to centimeter-scale away from the analyte. Such short distance minimizes interference from the external physical environment to give high signal-to-noise ratios. Thus, close-range sensing is widely applied in laboratory environment for high precision and quantitative SERS measurements. Herein, we focus on the development of 3D SERS platforms toward diversified sensing scenarios, including multiplex sensing and microfluidic high-throughput sensing.

Miniature 3D interfacial SERS platforms including plasmonic liquid marbles and plasmonic colloidosomes, are appealing for simultaneous interfacial and multiplex detections (Figure 6A).[35] In comparison to 2D monolayer interfacial SERS platforms, these micrometer-size 3D SERS platforms have excellent tolerance for laser misalignment while using ultrasmall sample volumes (μL or sub-μL), giving rise to detection limits down to sub-femtomole level.[36] Moreover, the nature of 3D capsule allows the encapsulation and storage of analyte molecules, hence enables molecular tracking in a mixture of multiple samples via 3D hyperspectral SERS imaging (Figure

6A). This allows the establishment of plasmonic colloidosomes as the first “dual-phase tri-analyte” detection system, whereby three analytes existing in two different phases are simultaneously detected. The analytes whereabouts are also traced and imaged in a 3D spatial volume.

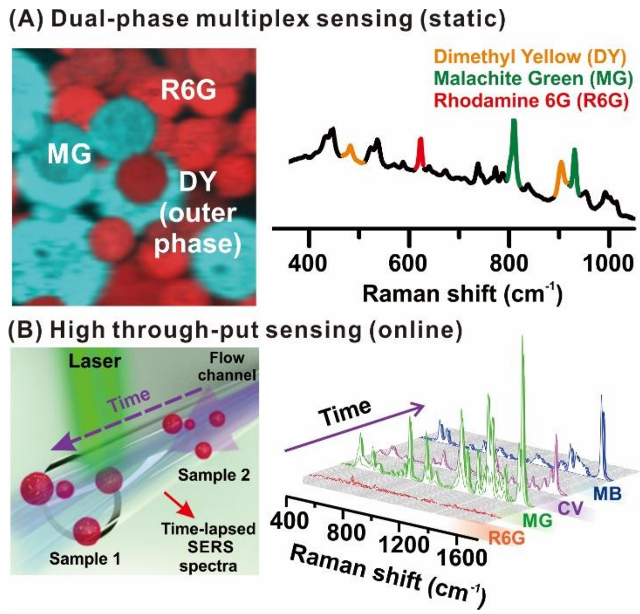

Figure 6. Close-range molecular sensing. (A) Dual-phase multiplex SERS sensing using 3D interfacial plasmonic colloidosomes and static hyperspectral x-y imaging revealing samples containing three analytes in two phases. Reproduced with permission from ref 36. Copyright 2016 John Wiley & Sons, Ltd. (B) On-line SERS detection using plasmonic colloidosome for sequential and quantitative multianalyte detection. Reproduced with permission from ref 42. Copyright 2017 John Wiley & Sons, Ltd.

Furthermore, we incorporate the plasmonic colloidosomes in a microfluidic channel, and realize on-line and rapid high through-put analysis of multiple samples (Figure 6B).[42] The analytes encapsulated inside 3D colloidosome remain isolated, thus preventing analyte cross-talk and channel contamination for sequential detection and allowing accurate quantification of samples over a concentration range of five orders of magnitude (10^{-7} to 10^{-2} M). The collective advantages provided by 3D interfacial SERS platforms promotes the development of portable sensors based on PLM and colloidosomes for applications in medical diagnosis, food sampling, and environmental monitoring.

4.1.2 Stand-off sensing

In contrast to close range sensing, stand-off sensing denotes the remote detection, identification and quantification of chemical compositions at long distances. It is typically employed to safeguard both personnel and instruments when the analyte is at dangerous or inaccessible locations. Stand-off Raman involves a telescope incorporated Raman spectrometer to collect analyte Raman signals at meter-scale distances. However, existing stand-off Raman spectroscopy systems suffer from intrinsically weak Raman scattering signals (only $1/10^6$ scattering photons),[2] and require high laser power (~400 mW) or long acquisition time (> 60 seconds),[43-45] making it an inefficient remote sensing tool.

To address these limitations, we incorporate SERS with stand-off Raman spectroscopy system to effectively amplify the Raman signals of remote probe analytes. Aerosolized plasmonic colloidosomes are coupled with stand-off Raman to form the very first substrate-less in-air stand-off SERS system. We demonstrate the detection of airborne pollutants at a distance of 200 cm, with detection limit of 5 nmol (Figure 7).[24] This aerosolized platform serves as an omnidirectional plasmonic cloud that enables sensitive detection from any direction and also eradicates the need for precise laser alignment. The APC can also perform multiplex detection upon exposure to a mixture of airborne analytes, revealing their signature fingerprint bands. In order to further propel the practical application of 3D aerosolized plasmonic colloidosomes in real world, the signal interference from the external environment, such as temperature, weather, and background light should be considered and improved. Additionally, we aim to develop in-air platforms with analyte-sorbing ability which is of great significance to improve the signal intensities and specificity even with complicated background matrices.

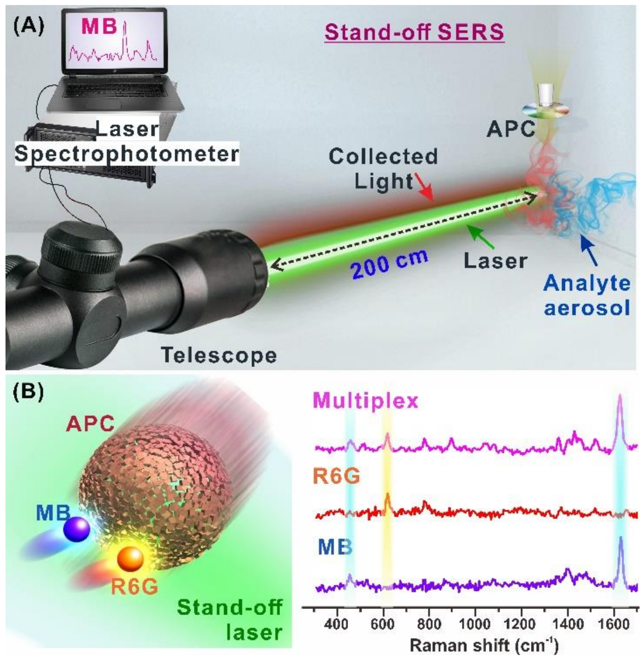

Figure 7. 3D SERS application in stand-off detection. (A) Scheme depicting the setup of stand-off SERS for detection of airborne analytes using aerosolized plasmonic colloidosomes (APCs). (C) Scheme and SERS spectra of in-air multiplex detection of both methylene blue and rhodamine 6G by APCs. The characteristic peaks of MB (456 and 1630 cm^{-1}) and R6G (620 cm^{-1}) are highlighted in blue and yellow, respectively. Reproduced with permission from ref 24. Copyright 2018 John Wiley & Sons, Ltd.

4.2 Plasmonic microlabs

Recently, SERS platforms have been combined with droplet-based microreactors for real-time assessment of chemical processes, critical for optimization of reaction yield, selectivity, and multi-step reaction pathway. The combination of 3D plasmonic liquid marble and plasmonic colloidosomes with SERS hyperspectral imaging creates excellent micro-testbeds for *in situ* reaction monitoring at molecular level through the 3D SERS-active plasmonic shell. Unlike *ex situ* techniques, we are able to track reaction events in real time without disturbing the reaction setup. This mitigates the potential harms arising from hazardous or explosive reactions. Using a 2-μL PLM, we uncover a two-step sequential pathway for diazonium-based moieties grafting onto Ag surface and elucidate the reaction kinetics for the first time. We also showcase the PLM as an ideal miniaturized chemical plant to conduct on-demand reaction-and-detection sequences (Figure 8A).[46] Utilizing a two-step azo-dye formation as our model reaction, our microchemical plant can merge and mix to achieve the rapid and efficient diazotization of nitroaniline to form diazonium nitrobenzene, followed by the azo coupling with aromatic compounds to yield azo-dye. Notably, we can also track the *in situ* molecular reactions in a non-disruptive manner. On the other hand, we establish the versatility of plasmonic colloidosomes and demonstrate the ability to *in situ* monitor multiple interfacial reactions at the pico-liter scale using SERS (Figure 8B).[23] For example, we successfully monitor and derive the reaction order and kinetics of dimethyl yellow protonation, and unprecedentedly differentiate the two isomeric products arising from the protonation at different sites.

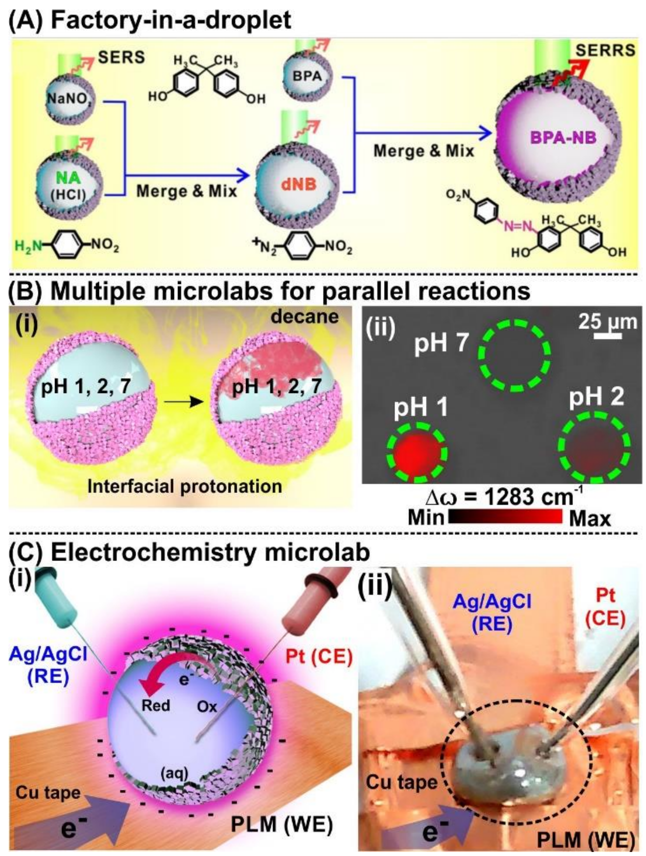

Figure 8. **Plasmonic microlabs.** (A) Scheme illustrating the use of PLM-based microchemical plant for sequential two-step synthesis of azo-dye and concurrent SERS monitoring. Adapted with permission from ref 46. Copyright 2017 American Chemical Society. (B)(i) Interfacial protonation of DY (DY—yellow) to form protonated DY (HDY^+—red) on colloidosomes encapsulating aqueous solution. (ii) SERS hyperspectral imaging reveals different reaction extent for colloidosomes encapsulating different pH solutions. Adapted with permission from ref 23. Copyright 2017 John Wiley & Sons, Ltd. (C)(i) Scheme illustrating the *in-situ* SERS detection on electrochemical reaction performed within a PLM, and the use of the Ag shell as 3D working electrode (WE). (ii) Digital image of the EC-SERS setup. Adapted with permission from ref 47. Copyright 2017 John Wiley & Sons, Ltd.

Moreover, PLM has been combined with electrochemistry (EC) for concurrent spectro- and electrochemical analysis in real time (Figure 8C).[47] The 3D Ag shell of PLMs can act as a bifunctional SERS platform and working electrode for redox process modulation which enable real-time molecular-level identification of transient species. Such spectro-electrochemical microliter scale reactor is crucial for elucidation of tandem EC reactions dynamics, which is impossible by standalone EC methods. Our finding is thus valuable to impact future design of 3D electrode system for enhanced electrochemical performance relevant in the broad applications in environmental conservation, energy harvesting, and biochemical charge transfer. These collective advantages and the versatile applications further highlight 3D plasmonic microlabs as advanced reactor-sensor hybrids that are beneficial for investigating multiple reactions and/or interfacial phenomena in many (bio)chemical processes.

4.3 Data storage and security labelling.

SERS-based informatics is an attractive and upcoming application of SERS, especially in the engineering of data storage and anti-counterfeiting platforms.[22] Using plasmonic micro- or nanostructures, information can be encoded within the structures via embedding probe molecules on them. The use of SERS further improves the security of the data and makes it much less duplicable, because the specificity of Raman fingerprinting allows the differentiation between spectroscopic features with resolution of 1 cm^{-1}. Therefore, this molecular information adds several security levels in both data encoding and decoding because knowledge and techniques far beyond simple physical analysis is required to decipher/mimic. This has resulted in the engineering of several SERS-based data storage and security labels that qualify for Level 3 security (L3S – refers to when encoded information can only be retrieved by advanced forensic tools with authorization), via the fabrication of 2D molecular-embedded plasmonic patterns with lithography techniques.

To achieve ≥ 4-fold higher storage density than 2D platforms, we develop 3D SERS anticounterfeiting platform by combining 3D candlestick microstructures with 3D hyperspectral SERS imaging (Figure 9).[22] By using microstructures of three different heights, we create three varying patterns that can be visualized at the corresponding z-focal plane. Thus, such 3D platform enables at least three tiers of encoded information within the same 2D area along the z-axis, significantly raising the difficulty in decoding/duplicating this information. Notably, the 3D security labels can be directly fabricated on both rigid and flexible substrates, widening their applicability. Our findings further highlight the tremendous potential of 3D SERS platform and provide valuable insight for future design of security label with enhanced security level for anti-counterfeiting application.

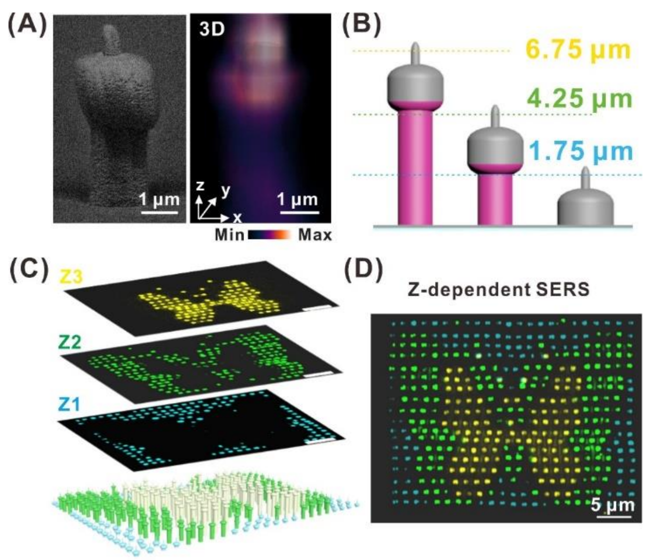

Figure 9. Data encryption and security label system using 3D SERS platforms. (A) Side-view SEM and 3D SERS images of a 3D candlestick microstructure. (B) Scheme showing three-layered Ag-coated candlesticks of different heights (1.75, 4.25 and 6.75 μm) that are arranged in pre-defined

pattern in the security label scheme in (C), which shows the SERS images obtained by adjusting the laser focal plane at different heights. (D) Overlap images of all the Z-dependent SERS images. Adapted with permission from ref 22. Copyright 2017 American Chemical Society.

5. OUTLOOK AND CONCLUSION

Since the discovery of SERS, it has first been developed as a Raman enhancing technique to push the limit of chemical detection, and then gradually as a robust analytical method that accompanies various nano- and microscopic platforms. The construction of SERS platforms in all three dimensions has further enabled the design of new microreactors, online analyzers, and even security labels that potentially serve to solve real-world issues. In this account, we have elaborated on our contribution to the frontier of 3D SERS platforms design and how it paves the way for trending applications of SERS in both detection and non-detection areas. While we discuss extensively about other applications beyond sensing, we recognize that SERS research can only be developed further when SERS as a sensing method is first strengthened. Indeed, SERS sensing still faces multiple challenges in detecting small molecules that have no affinity to metal surface such as hydrogen (H_2), which are not detectable even using 3D SERS platforms of strong SERS capabilities. It requires the parallel development of both SERS sensing capability of plasmonic NPs and 3D platform engineering to achieve new solutions for real-world problems. Hence, in addition to the assembly of nanoparticles into 3D structures, particle modifications for enhanced analyte capturing and sensitivity are crucial for both sensing purposes and reaction monitoring or optical storage. Nevertheless, the significant SERS applications we showcase are only achievable via 3D platforms, and this highlights the tremendous potential in providing breakthrough solutions for both infancy research and product engineering in various fields. Consequently, our motivation for this review is to summarize the latest advancement of SERS in its process to venture into interdisciplinary technologies and provide interested researchers with a comprehensive yet compact guide to learn about and build on such process. With this vision, we foresee the creation of highly applicable 3D SERS platforms that excel in both sensing and non-sensing areas which can further be developed into commercial products to solve technological challenges, improve global environmental conditions and ultimately human quality of life.

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