Chapter 15

Plasmonic Nanostructures as Surface-Enhanced Raman Scattering (SERS) Substrate for Protein Biomarker Sensing

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Additional information is available at the end of the chapter

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Abstract

Plasmonic nanostructures have attracted considerable interest in biomarker sensing with the goal of rapid diagnostics and personalized nanomedicine. Surface-enhanced Raman scattering (SERS) is a versatile technique for the characterization of the plasmonic effect of the metallic nanostructures as well as a sensitive read-out approach for biomarkers detection. In this contribution, we will give a review on the key optical properties of plasmonic nanostructures as SERS substrate for protein biomarkers detection. As a consequence, two approaches, label-free and SERS labels will be discussed in details for protein biomarkers sensing by using the plasmonic nanostructures as the substrate.

Keywords: plasmonic nanostructures, surface-enhanced Raman scattering (SERS), protein biomarker, label-free, SERS labels, sensing

1. Introduction

In order to understand the fundamental of plasmonic nanostructures and the related application, it is important to start with "plasmon", which was first named by Pines in the 1950s [1, 2]. As the valence electron collective oscillations resemble closely the electronic plasma oscillations observed in gaseous discharges, the term "plasmon" was thus used to describe the quantum of elementary excitation associated with this high-frequency collective motion [2]. Based on the definition, plasmon is very similar as photon, which is the quantum particle representing the elementary excitations, or modes, of the free electromagnetic field oscillations. To be simple, plasmon can be described in the classical picture as an oscillation of free electron density against the fixed positive ions in a metal [3, 4]. To visualize a plasma oscillation,

imagine a cube of metal is placed in an external electric field pointing to the right. Electrons will then move to the left side (uncovering positive ions on the right side) until they cancel the field inside the metal [5]. When the electric field is switched off, and the electrons move to the right, repelled by each other and attracted to the positive ions left bare on the right side. They oscillate back and forth at the plasma frequency until the energy is lost in some kind of resistance or damping [6–10]. Plasmons are a quantization of this kind of oscillation, which plays a large role in the optical properties of metals. Light of frequency below the plasma frequency is reflected, because the electrons in the metal screen the electric field of the light. Light of frequency above the plasma frequency is transmitted, because the electrons cannot respond fast enough to screen it [5]. In most metals, the plasma frequency is in the ultraviolet, making them shiny (reflective) in the visible range [8–11]. Some metals such as copper and gold having electronic interband transitions in the visible range, whereby specific light energies (colors) are absorbed, yield distinct colors.

Surface plasmons are those plasmons that are confined to surfaces and that interact strongly with light resulting in a polariton. They occur at the interface of a vacuum or material with a positive dielectric constant and a negative dielectric constant (usually a metal or doped dielectric) [5]. Plasmonics is related to the localization, guiding and manipulation of electromagnetic waves beyond the diffraction limit and down to the nanometer-length scale [12, 13]. The key component of plasmonics is a metal, which can support surface plasmon polariton (electromagnetic waves coupled to the collective oscillations of free electrons in the metal, plasmon coupled with photons). There are two types of surface plasmon resonances (SPRs) that can be generated [12, 13]. The first one is the surface plasmon polariton (SPP), which is generated when light becomes trapped at a metal-dielectric interface. The second one is the localized surface plasmon resonance (LSPR), which happened when a surface plasmon is confined to a particle of size comparable to the wavelength of light, that is, a nanoparticle, the particle's free electrons participate in the collective oscillation. For the full detailed description of the fundamentals of plasmon and plasmonic effect, several excellent reviews are recommended [13–19].

Plasmonic nanostructures are thus the metallic nanostructures on which the electromagnetic field was generated by exciting the oscillation of surface plasmon in the metal-light dielectric interface. The study on the plasmonic nanostructures has attracted more and more interest in the research areas from the fundamentals to applications in a variety of scientific disciplines. In this chapter, we will focus on the plasmonic nanostructures as the surface-enhanced Raman scattering (SERS) substrate for protein biomarkers detection.

SERS was first discovered in the 1970s [20, 21] on electrochemical roughed silver electrode and dramatically developed with the advancement of nanotechnology. Currently, two basic principles that contribute the SERS effect are accepted, which are the chemical effect (CM) e.g., charge-transfer and the electromagnetic effect (EM) [22–28]. Basically, CM is the interaction of the adsorbate molecules and the metal surface, mostly from the first layer of the charge-transfer resonance between molecules and the metal [22–24]. Whereas EM mechanism is based on the interaction of the transition moment of an adsorbed molecule with the electric field of a surface plasmon induced by the incoming light at the metal, as discussed above [25–28].

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Thus, SERS has been utilized in different areas including the catalysis, energy, and biology, and, in particular, in biomedical application. For instance, SERS has been used to monitor the catalytic reaction, image the live cell and tumor, monitor the nanoparticle distribution in live body, and so on [29–31]. Several excellent reviews have been published on SERS including the basic principle of SERS, SERS nanoparticles, SERS labels as well as SERS applications in biomedicine [32–37]. In this chapter, we will mostly focus on the optical properties (plasmonic effect) of the metallic nanostructures for SERS, with the highlight on the application of plasmonic nanostructures for protein detection by using SERS as a read-out technology.

2. Plasmonic nanostructures

In this section, optical property of plasmonic nanostructures (surface plasmon resonance) will be discussed in terms of the SERS-activity. Plenty of methods have been reported for the synthesis of plasmonic nanostructures including the wet-chemistry (seed-mediated growth), template nanoscale lithography, thin-film, and the template approach. Here in this chapter, we will mainly focus on the synthesis and characterization of plasmonic nanostructures that are widely used for SERS study. Based on the shape or geometry of the nanostructures, we will give a brief overview for the quasi-spherical metallic nanostructures, anisotropic nanoparticles/structures as well as the plasmonic nanoassemblies with the highlight on the plasmonic effect for SERS activity. As Mirkin et al. have discussed in details of the template techniques for assembly of plasmonic nanostructures [13], we will not include this technique in this chapter.

2.1. Quasi-spherical metallic nanoparticles

As gold and silver nanostructures show the most significant SERS effect due to the surface plasmon resonance generated on gold and silver surface (EM enhancement mechanism), other metals such as copper and the transit metal also show the SERS effect; however, the impact factor is very low, which depends on several factors, including the size and geometry of nanoparticles as well as the laser excitations. Numerous protocols are available for the synthesis of metallic spheres [38–41]. The simplest and most common approach is the reduction of metal salts with a variety of reducing and capping agents. To improve the stability and the enhancement of plasmonic nanoparticles, the combination of the alloy metal is an option. Thus, gold/silver nanoshells are designed and synthesized as the composite metallic nanostructures [42, 43] that show higher SERS effect compared to single metal. More importantly, by tuning the shell thickness, the LSPR of gold/silver nanoshells can be tuned (**Figure 1b**). In regard to the enhancement factor (EF) on the metallic sphere nanoparticles for SERS, it has been reported that individual spherical nanoparticles generate very low EF for SERS based on the EM calculation [34, 44]. Single molecule detection [25, 27] was reported on silver nanoparticles with the enhancement factor (EF) around 10^{12}–10^{14}, which is due to the strong surface plasmon coupling effect (called "hot-spot"). In order to get high order of EF, well-defined plasmonic nanostructures with multiple "hot-spots" are required, which will be discussed in the following session.

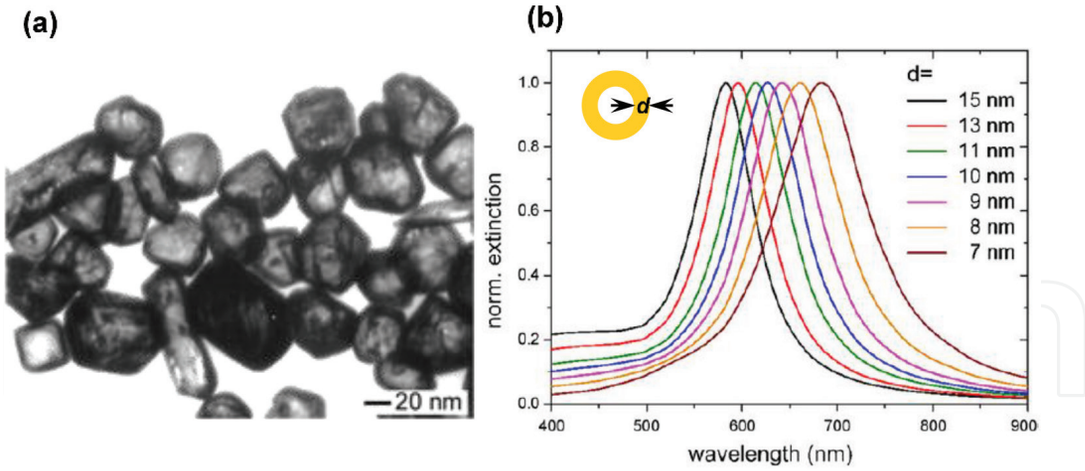

Figure 1. TEM image of gold/silver nanoshell (a) and the tunable LSPR of gold/silver nanoshells with the shell thickness (b) (cited from Ref. [33] with permission from the Royal Society of Chemistry).

2.2. Anisotropic nanoparticles

As it is hard to obtain the SERS signal in the individual metallic nanospheres, anisotropic nanoparticles such as rods, stars, cubes, prisms, and nanoplates became very important SERS substrates (**Figure 2a**), which exhibit significantly higher electromagnetic field enhancements at sharp edges ("lighting rod effect" or "plasmonic antenna effect"), making them attractive for use as plasmonic enhancers in SERS [45–49]. For the synthesis of anisotropic nanoparticles, the most widely employed approach is seed-mediated growth, which involves seed-formation and growth. Typical example is the synthesis of gold nanorods (**Figure 2a**), which starts with the quasi-spherical ~4 nm gold seeds and subsequent reduction of more metal salt with

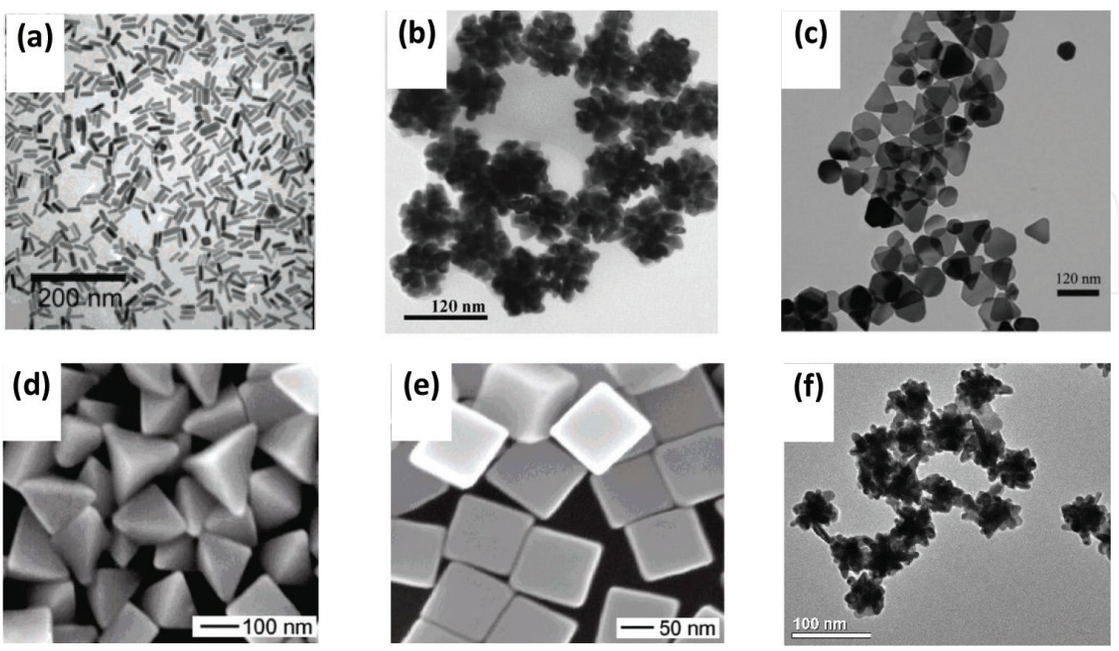

Figure 2. Typical electronmicrographs of anisotropic nanostructures for efficient SERS. Nanorods (a), nanoflowers (b), nanoplates (c), nanoprisms (d), nanocubes (e), and nanostars (f). (cited from Ref. [33] with permission from the Royal Society of Chemistry).

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a weak reducing agent such as ascorbic acid in the presence of structure-directing additives e.g., CTAB (cetyltrimethylammonium bromide), leading to the formation of gold nanorods. The aspect ratio can be controlled by the relative concentrations of the reagents. By tuning the aspect ratio of the gold nanorods, their localized surface plasmon resonance (LSPR) can be tuned, which is an important aspect for the application of gold nanorods in SERS as well as phototherapy.

2.3. Plasmonic nanoassemblies

In order to generate the hot-spots for the SERS enhancement, plasmonic clusters including the dimers, trimers, and high orders nanoaggregates have been prepared. As it is very important to understand the relationship between nanoparticle structure and the SERS activity, Van Duyne et al. have reported the SERS EF with the different nanostructures (mainly dimers and trimers) [50]. As indicated in **Figure 3**, individual trimers have been encapsulated with silica-shell to avoid the interference from the environment as well as ensure the contribution solely from the individual nanoparticles. It was demonstrated that the creation of hot spots, where two nanoparticles are in sub-nanometer proximity or have coalesced to form crevices, is the paramount to achieving maximum SERS EF. Specifically, L-shaped trimer nanoantenna comprised of three Au cores showed the EF of 10^8–10^{10} (**Figure 3a**) and the dark-field Rayleigh scattering spectrum of the L-shaped trimer contains three peaks (**Figure 3b**), corresponding to dipolar and multipolar LSPRs [50]. Single-particle SERS obtained from the trimer nanoantenna showed distinct peaks from the Raman molecule (PCEPE), which are correlated with the density functional theory (DFT) calculations (**Figure 3c–e**).

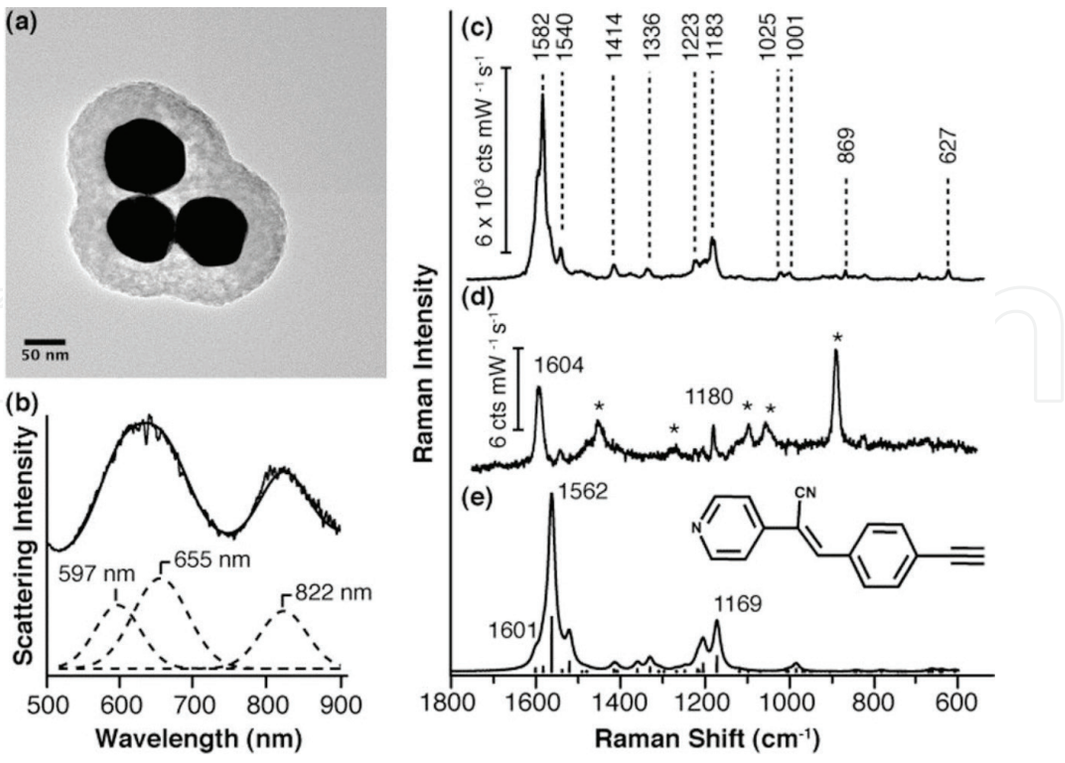

Figure 3. L-shape trimer nanoantenna (a), LSPR spectrum of the trimer obtained by dark-field Raleigh scattering microscopy (b), SERS spectrum of the Raman molecule (PCEPE) (c), normal Raman spectrum of PCEPE (d), and Raman spectrum of PECEP calculated using DFT (e). (Reproduced with permission cited from Ref. [50]).

Plasmonic nanostructures with multiple hot-spots have been reported with the satellite nanostructures by using either gold sphere or gold nanorods as the core [51–53]. To obtain the high orders of nanoassemblies, the linker is the key to connect the nanoparticles. Chemicals with dual/multifunctional groups are often used such as cystamine and dithiol-polymer. As silica surface is versatile, the functionalization of silica surface followed by the assembly of small nanoparticles is a very useful strategy. More importantly, the distance between the satellite nanoparticles with the core nanoparticles can be controlled by the silica-thickness. As reported by Gellner et al. [51], gold nanoparticles with the diameter of 80 nm were incubated with Raman reporters and encapsulated with a very thin silica shell. The ultrathin glass shell was then functionalized with a binary mixture of silanes including an aminosilane, which can adsorb the negatively charged gold nanospheres to form the 3D structures. Correlated HR-SEM/dark-field/LSPR/SERS experiments on individual 3D SERS-active superstructures together with finite element method (FEM) calculations confirmed the plasmonic coupling between the core and the satellite particles, with hot-spots occurring between core and satellites as well as between satellites [51].

3. Label-free protein biomarkers detection

SERS is a powerful vibrational spectroscopy, which can provide rich molecular information for the target, making it very useful for direct protein detection. The fingerprint information extracted from the SERS spectra of proteins can be used directly to identifying the protein confirmation, the structure as well as the component of the target protein [54–56]. Label-free SERS detection for protein originates from the chromophores such as hemoglobin, myoglobin, and cytochrome c, which showed strong SERS signals with good reproducibility due to the Raman resonance (RR) effect of the chromophore center of the proteins [57, 58]. The information related to the conformation and orientation of proteins as well as the charge-transfer processes between protein and surface can be obtained. As reported by Feng and Tachikawa [59], to determine the factors that contribute to the difference of SER(R)S signals and RR signal of metmyoglobin, they designed a Raman flow system and found that both the degree of protein-nanoparticles interaction and the laser irradiation contribute to the structural changes (**Figure 4**).

As the majority of proteins have no chromophore, the detection of proteins become much harder since the SERS signal from the native protein is very weak and most of the signals are generated from the amino acid residues and amide backbones, which are very similar for most of the proteins [60, 61]. Therefore, the key to get robust and sensitive label-free protein detection is the SERS substrate, which should have the high SERS-activity as well as proper preparation of the reproducible surface. Gold and silver nanocolloids are the widely used plasmonic nanoparticles for the label-free SERS detection [62–64]. As the gold and silver nanoparticles have relatively lower SERS enhancement effect, the procedure that could induce the aggregation, thus generate "hot-spot" nanostructures is required. Typically, the addition of aggregation agent such as salt is the simplest and easiest approach for the aggregation. As displayed in **Figure 5**, Han et al. utilized sulfate as an aggregation agent to induce strong

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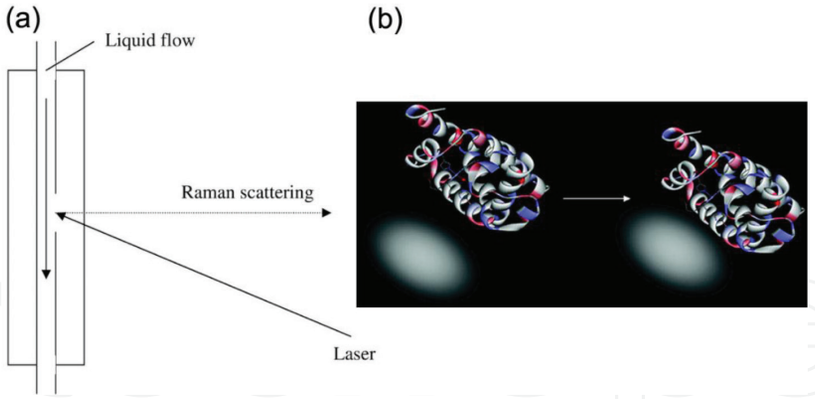

Figure 4. Raman-flow system for label-free protein detection (a) and the interaction of the protein with a silver nanoparticle monitored by SERS (b). (Reproduced with permission cited from Ref. [59]).

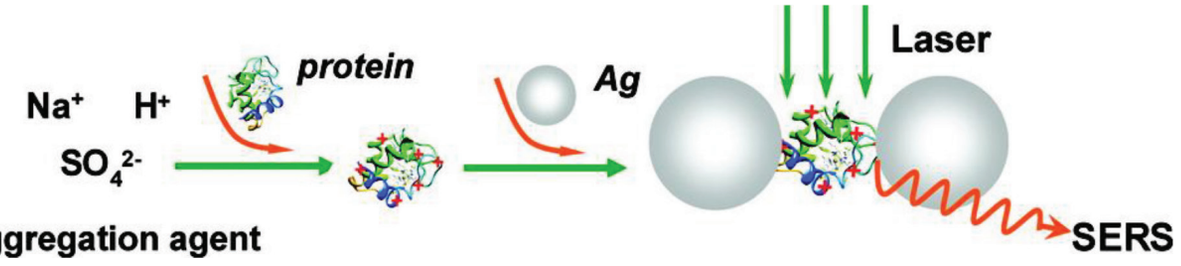

Figure 5. Scheme for aggregating nanoparticles for directly label-free SERS detection of proteins. (Reproduced with permission cited from Ref. [65]).

SERS signal due to the weak binding of SO_4^{2-} on silver surface, making it easier for protein binding [65]. With this scheme, proteins including lysozyme, ribonuclease B, avidin, catalase, and hemoglobin have been detected and analyzed [65]. As SERS signal from the protein can provide useful information in terms of the structure, constituents, conformation as well as the potential interaction of the protein with the surface, in recent years, several groups have put great effort to improve the method of SERS-based label free identification of proteins. For instance, Zhao et al. have conducted serious study on the label-free protein detection by controlling the plasmonic nanoparticles and optimizing the purification procedures of the proteins [62–65]. A typical example is the Western-blot SERS, which was based on the silver staining of the membrane after the protein separation with the gel electrophoresis, which will purify and separate the proteins on the surface, making the detection more simple and easy.

Furthermore, Ren et al. have proposed a facile method to enable reliable label-free SERS detection of the native structures of a wider range of proteins by using the iodide-modified silver colloids as illustrated in **Figure 6** [66]. The colloidal state of Ag NPs will help to keep the native structures of proteins and promote the photostability of samples. The iodide modification affords a one-atom-thick monolayer on the surface without producing interfering signal. Therefore, they demonstrated that the iodide-modified silver colloids could not only clean the surface but also avoid the strong chemical interaction between the metal surface and the proteins, and reduce the possibility of denaturation, thus make the detection reliable and reproducible.

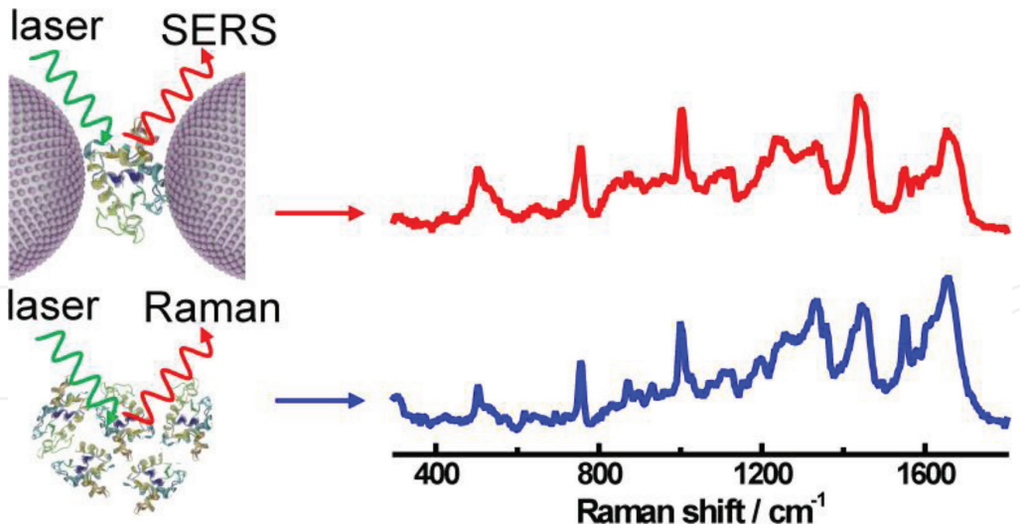

Figure 6. Label-free SERS for native protein detection on iodide-modified silver nanoparticles. (Reproduced with permission cited from Ref. [66]).

Recently, label-free SERS has been attempted for the serum protein detection, aiming for the cancer diagnosis. By inducing the aggregation of silver nanoparticles for high-quality SERS spectra, colorectal cancer has been identified based on the principal component analysis combined with linear discriminant analysis. [67–69]

4. SERS Labels for protein biomarkers sensing

Compared to label-free SERS approach, SERS labels have attracted significant attention for protein detection because of its high sensitivity comparable to fluorescence. The high sensitivity is most due to the strong plasmonic effect of the SERS substrates, which could be AuNPs, AgNPs, anisotropic nanoparticles (gold nanorods), nanoshells, plasmonic nanoassemblies, as discussed in the early session of this chapter. More importantly, SERS labels have demonstrated their unique optical properties and potential for simultaneous and multiplexed detection [33–37] owing to the advantages of SERS labels over fluorescent label including (i) the multiplexing capability for simultaneous target protein detection due to the narrow width of the vibrational Raman bands, (ii) quantification using the SERS fingerprint of the corresponding labels, (iii) the need for only a single laser excitation wavelength, and (iv) high photostability. In this section, the sensor platform using SERS labels for protein biomarker sensing will be discussed from the conventional sandwich immunoassay, dot-blot semi-sandwich immunoassay, and protein microarray to microfluidic protein assay, with the highlights on our recent works for the detection of breast cancer biomarker [70–73], pathogen antigens [74], cytokines [75, 76], and related works.

SERS sandwich immunoassay is a conventional sensor platform for the protein biomarker detection with SERS labels. Porter et al. have reported a serial of works on SERS immunoassay for various protein biomarkers detection [77–82]. The SERS labels were named as ERLs (external reporter label). Typically, ERLs were composed of metallic nanoparticles (e.g., widely

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used gold nanoparticles duo to the distinguished plasmonic effects) and Raman reporters (dyes or the small molecule) to indicate the presence of the target. As indicated in **Figure 7a**, detection antibodies were immobilized on the plasmonic nanoparticles by either electrostatic force or the covalent binding through the links such as DSP (dithiobis (succinimidyl propionate)) [79] and carboxyl-PEG-SH (poly (ethylene glycol) 2-mercaptoethyl ether acetic acid) [30]. To fabricate the sandwich SERS platform, the substrate could be the glass or gold film (**Figure 7b**). Additional studies have shown the higher sensitivity with gold film as the substrate because of the plasmonic coupling effect between the gold film (SPP—surface plasmon polarition) and the SERS labels (LSP—localized surface plasmon) [81, 82]. Followed by the capture of the target antigen or probes by the antibody on the surface, ERLs are bound on the surface for the signal to indicate the presence of the target proteins. With the similar platform design, plenty of proteins have been detected with few from the clinic important biomarkers [78–83].

Microassay is the platform that could detect proteins with high throughput by SERS labels using different dyes as the Raman reporters. As a typical example indicated in **Figure 8**, multiple protein targets have been immobilized on the surface, followed by the probe with the dye labeled SERS nanoparticles [83]. Upon the laser excitation, SERS signals from the dye will indicate the presence of the target proteins. Microfluidic assay is another platform with high-throughput properties for protein detection as the design for the channels can separate each of the individual proteins with different channels as well as enhance the reaction by controlling the flow conditions [74].

Although it is a high throughput, the requirement for the professional training, long-time incubation as well as the labor intensive procedures has hindered the application of the platform in the clinic setting. To improve the efficiency of the platform, a dot-bot assay was thus developed, which is targeting on the rapid and sensitive and simultaneous multiple protein

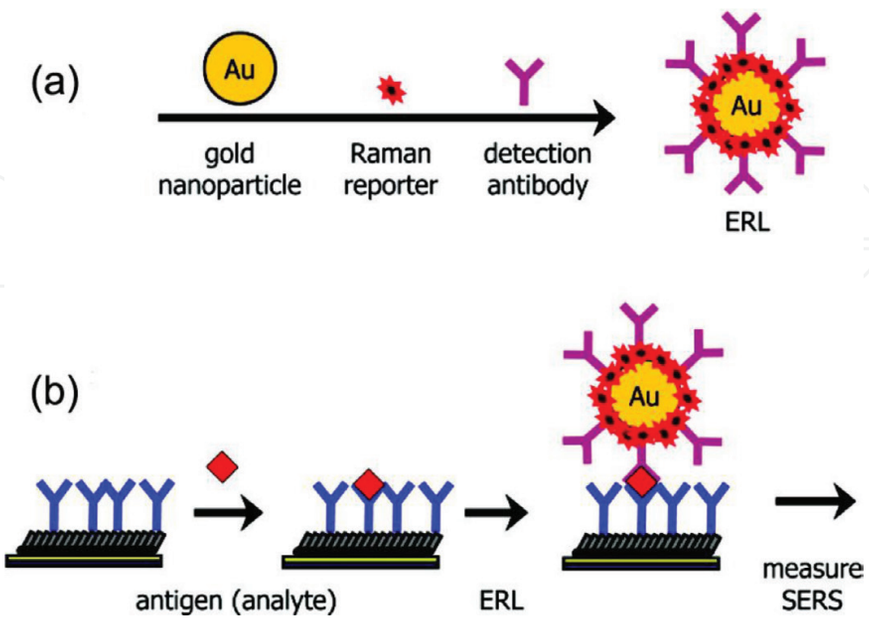

Figure 7. Schematic illustration of SERS ERLS (a) and typical SERS sandwich immunoassay for protein biomarkers detection (b). (Reproduced with permission cited from Ref. [35]).

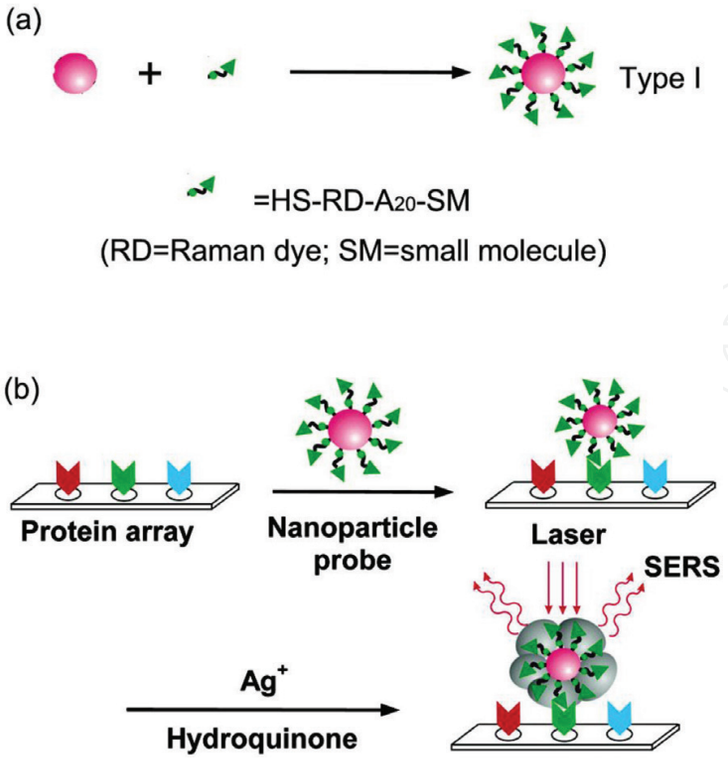

Figure 8. Dye labeled SERS nanoparticles (a) and protein array (b) for high throughput proteins detection by SERS. (Reproduced with permission cited from Ref. [83]).

detections with a simplified procedure [75]. As displayed in **Figure 9**, duplex cytokines (interleuckin-6 and interleuckin-8; IL-6 and IL-8) were detected simultaneously on the dot-blot assay with femtogram (fg) sensitivity, which was achieved by using the rational designed Au/Ag nanoshells as the plasmonic substrate (**Figure 9**). As specificity is a key issue for this study because it is hard to test the real samples without purification. Therefore, much more works were reported in this area, the limitations in the long incubation time and the labor-intensive procedures with this platform have become a big obstacle toward the application in real, in particular, for the point-of-care diagnosis.

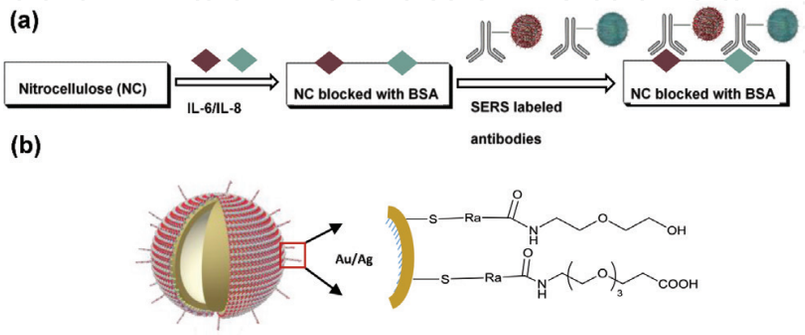

Figure 9. Scheme of the direct SERS dot-blot immunoassay platform for duplex cytokine detection (a), schematic illustration of hydrophilically stabilized Au -Ag nanoshells with Ra-MEG-OH/TEG-COOH (b). (cited from Ref. [75] with permission from the Royal Society of Chemistry).

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To minimize the assay time and enable the rapid detection, several strategies have been employed [80, 84]. For instance, Driskell et al. prepared an approach to increase the flux of antigen and SERS particles to the solid-phase surface by using a rotated capture substrate. As illustrated in **Figure 10a**, by controlling the rotating rate for the capture substrate, the reaction kinetics can be improved quickly [80]. Instead of gold nanoparticles, gold nanorods were used as plasmonic nanostructures for ERLs to improve the sensitivity. The assay time was reduced from 24 hours to 25 minutes, however, in a 10-fold loss of sensitivity compared to the conventional SERS sandwich immunoassay. Further, to improve the simplicity of the assay, the syringe pump SERS immunoassay platform (**Figure 10b**) was developed to overcome diffusion-limited binding kinetics that often impedes rapid analysis in conventional SERS immunoassay. The assay time was reduced from 24 hours to 10 minutes with a 10-fold improvement in detection limit [84].

Despite these attempts being successful in reducing assay times, nonspecific adsorption of nontarget molecules still remains the biggest challenge in immunoassay for protein detection. To circumvent this problem, our group recently proposed an innovative platform that utilizes nanoscaled alternative current electrohydrodynamic (ac-EHD)-induced surface shear forces to enhance the capture efficiency as well as significantly reduce the nonspecific binding of the molecules on the surface (**Figure 11a**). Meanwhile, to improve the sensitivity, rational designed silica-coated gold/silver nanoshells have been employed as the SERS labels. It was found that the detection limit can go down to 1 fg/mL. Further, to improve the design of the channel (**Figure 11b**), simultaneous detection of four biomarkers was achieved both from the serum and patient samples.

Due to the great advantages of SERS in the sensitivity, multiplexed capability with only one single laser excitation, and photostability, it is expected that SERS labels will have more applications in the point-of-care diagnosis platforms for protein biomarkers detection by using the rationally designed plasmonic nanostructures.

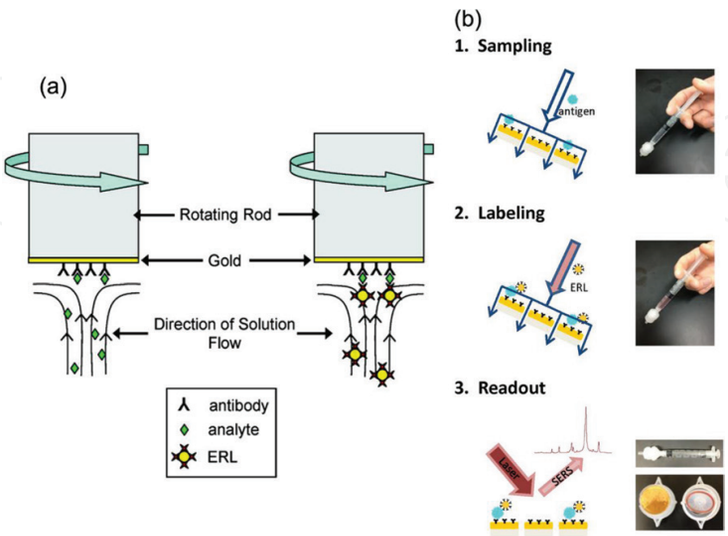

Figure 10. Two typical platforms of rotating capture substrate (a) and syringe pump (b) to enhance the assay time (Reproduced with permission cited from Refs. [80, 84]).

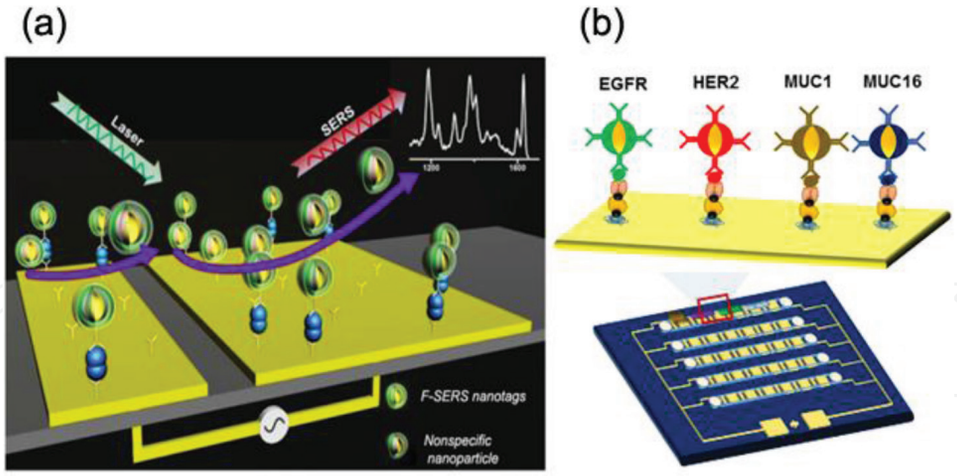

Figure 11. Scheme of the microfluidic device platform for protein biomarker sensing by using ac-EHD to enhance the assay time and minimum the nonspecific binding by applying a potential on the unsymmetry electrode pair. Dual functional Au/Ag nanoshells were used as SERS nanotags for breast cancer biomarker, HER2 detection (a) and simultaneously detecting four biomarkers in a five-channel device (b). (Reproduced with permission cited from Refs. [70, 71]).

5. Conclusions and perspectives

Plasmonic nanostructures with various size and shapes have been utilized in different areas including the catalysis, energy, and biomedicine. In this chapter, the fundamentals of plasmonic effect as well as the plasmonic nanostructures have been reviewed. Surface-enhanced Raman scattering (SERS) is an optical phenomena happened on the plasmonic nanostructures and have shown distinguished properties for protein biomarkers detection. With two typical approaches (label-free and SERS label) developed, SERS with plasmonic nanostructures has shown great potentials for proteins detection further to understand the biological system that protein involved. Meanwhile, although SERS detection for the protein biomarkers has been reported since 1990s, the application of this technology toward the real clinical sample is limited due to the rich information on the Raman peaks from the proteins, which have similar chemical bonds, thus making it very hard to be identified quickly and easily. Thanks to the development of the approach for the statistical analysis, label-free SERS detection for proteins has become feasible for clinical samples. On the contrast, SERS labels with the well-designed plasmonic nanoparticles show great potential for sensitive, reproducible, and simultaneous multiplexed detection for critical protein biomarkers. By combining the label-free SERS with SERS labels approach, we expect that SERS combined with the rational designed plasmonic nanostructures will greatly enhance the research and application in this field.

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References

[1] Pines D, Bohm DA. Collective description of electron interactions: II. Collective vs individual particle aspects of the interactions. Physical Review. 1952;**85**:338

[2] Pines D. Collective energy losses in solids. Review of Modern Physics. 1956;**28**:184

[3] Le Ru EC, Etchegoin PG. Principles of surface-enhanced Raman spectroscopy and related plasmonic effects. 2008, ISBN: 978-0-444-52779-0

[4] Schlücker S. Surface enhanced Raman spectroscopy: Analytical, biophysical and life science applications. 2011, ISBN: 9783527325672, DOI: 10.1002/9783527632756

[5] https://en.wikipedia.org/wiki/Plasmon

[6] Echenique JM, Pitarke, Silkin VM, Chulkov EV, Echenique PM. Surface plasmons in metallic structures. Journal of Optics A: Pure and Applied Optics 2005;**7**:S73-S84

[7] Raether H. Surface Plasmons on Smooth and Rough Surfaces and on Gratings. Springer Tracts in Modern Physics; 1988

[8] Maier SA. Plasmonics: Fundamentals and Applications. Springer: New York; 2007

[9] Kerker M. The Scattering of Light and Other Electromagnetic Radiation. Academic Press: New York; 1969

[10] Bohren CF, Huffman DR. Absorption and Scattering of Light by Small Particles. Wiley Interscience: New York; 1983

[11] Kreibig U, Vollmer M. Optical Properties of Metal Clusters. Springer: Berlin; 1995

[12] Willet KA, Van Duyne RP. Localized surface plasmon resonance spectroscopy and sensing. Annual Review Physical Chemistry. 2007;**58**:267-297

[13] Jones MR., Osberg KD, Macfarlane RJ, Langille MR, Mirkin CA. Templated techniques for the synthesis and assembly of plasmonic nanostructures. Chemical Review. 2011;**111**:3736-3827

[14] Morton SM, Silverstein DW, Jensen L. Theoretical studies of plasmonics using electronic structure methods. Chemical Review. 2011;**111**:3962-3994

[15] Giannini V, Fernandez-Dominguez AI, Heck SC, Maier SA. Plasmonic nanoantennas: Fundamentals and their use in controlling the radiative properties of nanoemitters. Chemical Review. 2011;**111**:3888-3912

[16] Mayer KM, Hafner JH. Localized surface plasmon resonance sensors. Chemical Review. 2011;**111**:3828-3857

[17] Rycenga M, Cobley CM, Zeng J, Li WG., Moran CEH, Zhang Q, Qin D, Xia Y. Controlling the synthesis and assembly of silver nanostructures for plasmonic applications. Chemical Review. 2011;**111**:3669-3712

[18] Halas NJ, Lal S, Chang W, Link S, Nordlander P. Plasmons in strongly coupled metallic nanostructures. Chemical Review. 2011;**111**:3913-3961

[19] Cortie MB, McDonagh AM. Synthesis and optical properties of hybrid and alloy plasmonic nanoparticles. Chemical Review. 2011;**111**:3713-3735

[20] Fleischmann M, Hendra PJ, McQuillan AJ. Raman spectra of pyridine adsorbed at a silver electrode original research article. Chemical Physical Letter. 1974;**26**:163-166

[21] Jeanmaire DL, Van Duyne RP. Surface Raman spectroelectrochemistry: Part I. Heterocyclic, aromatic, and aliphatic amines adsorbed on the anodized silver electrode. Journal Electroanalytical Chemistry and Interfacial Electrochemistry. 1977;**84**:1-20

[22] Ueba H. Theory of charge transfer excitation in surface enhanced Raman scattering original research article. Surface Science. 1983;**131**:347-366

[23] Otto A, Billmann J, Eickmans J, Erturk U, Pettenkofer C. The "adatom model" of SERS (Surface Enhanced Raman Scattering): The present status. Surface Science. 1984;**138**:319-338

[24] Arenas JF, Woolley MS, Otero JC, Marcos JI. Charge-transfer processes in surface-enhanced Raman scattering. Franck−Condon Active Vibrations of Pyrazine. Journal of Physical Chemistry. 1996;**100**:3199-3206

[25] Nie S, Emory SR. Probing single molecules and single nanoparticles by surface-enhanced Raman scattering. Science. 1997;**275**:1102-1106

[26] Doering WE, Nie SM. Single-molecule and single-nanoparticle SERS: Examining the roles of surface active sites and chemical enhancement. Journal of Physical Chemistry B. 2002;**106**:311-317

[27] Kneipp K, Wang Y, Kneipp H, Perelman LT, Itzkan I, Dasari RR, Feld MS. Single molecule detection using surface-enhanced Raman scattering (SERS). Review Letter 1997;**78**:1667

[28] Kneipp K, Kneipp H, Itzkan I, Dasari R, Feld MS. Ultrasensitive chemical analysis by Raman spectroscopy. Chemical Review. 1999;**99**:2957-2976

[29] Xie W, Schlucker S. Synthesis of bifunctional Au/Pt/Au Core/Shell nanoraspberries for in Situ SERS monitoring of platinum-catalyzed reactions. Journal of American Chemical Society. 2011;**133**:19302-19305

[30] Qian X, Peng XH, Ansari DO, Yin-Goen Q, Chen GZ, Shin DM, Yang L. In vivo tumor targeting and spectroscopic detection with surface-enhanced Raman nanoparticle tags. Nature Biotech. 2008;**26**:83-90

[31] Wang YL, Seebald JL, Szeto DP, Irudayaraj J. Biocompatibility and biodistribution of surface-enhanced Raman scattering nanoprobes in zebrafish embryos: In vivo and multiplex imaging. ACS Nano. 2010;**4**:4039-4053

[32] Wang YL, Wang EK. Nanoparticle SERS substrates, in Surface Enhanced Raman Spectroscopy: Analytical, Biophysical and Life Science Applications. Wiley-VCH; 2010;Chap. **2**:39-69

[33] Wang YL, Schlücker S. Rational design and synthesis of SERS labels. Analyst. 2013;**138**: 2224-2238

[34] Schlücker S. Surface-enhanced Raman spectroscopy: Concepts and chemical applications. Angewandte Chemie International Edition. 2014;**53**:4756-4795

[35] Porter MD, Lipert RJ, Siperko LM, Wang G, Narayanan R. SERS as a bioassay platform: Fundamentals, design, and applications. Chemical Society Review 2008;**37**:1001-1011

[36] Wang YQ, Yan B, Chen LX. SERS Tags: Novel Optical Nanoprobes for Bioanalysis, SERS nanotags for biomedical application. Chemical Review. 2013;**113**:1391-1428

[37] Lane LA, Qian X, Nie S. SERS nanoparticles in medicine: From label-free detection to spectroscopic tagging, Chemical Review, 2015;**115**:10489-10529

[38] Zhang Q, Xie J, Yu Y, Lee JY. Monodispersity control in the synthesis of monometallic and bimetallic quasispherical gold and silver nanoparticles. Nanoscale. 2010;**2**:1962-1975

[39] Evanoff DD Jr, Chumanov G. Synthesis and optical properties of silver nanoparticles and arrays. ChemPhysChem. 2005;**6**:1221-1231

[40] Daniel MC, Astruc D, Gold nanoparticles: assembly, supramolecular chemistry, quantum-size-related properties, and applications toward biology, catalysis and nanotechnology, Chemical Review. 2004;**104**:293-346

[41] Steinigeweg D, Schlücker S. Monodispersity and size control in the synthesis of 20-100 nm quasi-spherical silver nanoparticles by citrate and ascorbic acid reduction in glycerol-water mixtures. Chemical Communications. 2012;**48**:8682-8684

[42] Sun YG, Mayers BT, Xia YN. Template-engaged replacement reaction: A one-step approach to the large-scale synthesis of metal nanostructures with hollow interiors. Nano Letters. 2002;**25**:481-485

[43] Gellner M, Küstner B, Schlücker S. Optical properties and SERS efficiency of tuneable gold/silver nanoshells. Vibrational Spectroscopy. 2009;**50**:43-47

[44] Zhang Y, Walkenfort B, Yoon JH, Schlücker S, Xie W, Gold and silver nanoparticle monomers are non-SERS-active: a negative experimental study with silica-encapsulated Raman-reporter-coated metal colloids. Physical Chemistry Chemical Physics. 2015;**17**:21120-21126

[45] Murphy CJ, Sau TK, Gole AM, Orendorff CJ, Gao J, Gou L, Hunyadi SE, Li T. Anisotropic metal nanoparticles: Synthesis, assembly, and optical applications. Journal of Physical Chemistry B. 2005;**109**:13857-13870

[46] Xia Y, Xiong Y, Lim B, Skrabalak SE. Shape controlled synthesis of metal nanocrystals: simple chemistry meets complex physics. Angewandte Chemie International Edition. 2009;**48**:60-103

[47] Wiley BJ, Im SH, Li ZY, McLellan J, Siekkinen A, Xia Y. Maneuvering the surface plasmon resonance of silver nanostructures through shape-controlled synthesis. Journal of Physical Chemistry B. 2006;**110**:15666-15675

[48] Zou XQ, Ying EB, Dong SJ. Seed-mediated synthesis of branched gold nanoparticles with the assistance of citrate and their surface-enhanced Raman scattering properties. Nanotechnology. 2006;**17**:4758-4764

[49] Schütz M, Steinigeweg D, Salehi M, Kömpeb K, Schlücker S. Hydrophilically stabilized gold nanostars as SERS labels for tissue imaging of the tumor suppressor p63 by immuno-SERS microscopy. Chemical Commununication. 2011;**47**:4216-4218

[50] Wustholz KL, Henry AI, McMahon JM, Freeman RG, Valley N, Piotti ME, Natan MJ, Schatz GC, Van Duyne RP. Structure−activity relationships in gold nanoparticle dimers and trimers for surface-enhanced Raman spectroscopy. Journal of American Chemical Society. 2010;**132**:10903-10910

[51] Gellner M, Steinigeweg D, Ichilmann S, Salehi M, Schütz M, Kömpe K, Haase M, Schlücker S, 3D self-assembled plasmonic superstructures of gold nanospheres: Synthesis and characterization at the single-particle level. Small. 2011;**7**:3445-3451

[52] Choi I, Don H, Suseung S, YangYL, Kang T, Yi J. Core–satellites assembly of silver nanoparticles on a single gold nanoparticle via metal ion-mediated complex. Journal of American Chemical Society. 2012;**134**:12083-12090

[53] Fan Z, Tebbe M, Fery A, Agarwal S, Greiner A. Assembly of gold nanoparticles on gold nanorods using functionalized poly (N-isopropylacrylamide) as polymeric "glue". Particle & Particle Systems Characterization. 2016;**33**:698-702

[54] Deisenhofer J, Epp O, Miki K, Huber R, Michel H, X-ray structure analysis of a membrane protein complex. Electron density map at 3 A resolution and a model of the chromophores of the photosynthetic reaction center from Rhodopseudomonas viridis. Journal of Molecular Biology. 1984;**180** :385-398

[55] Picot D, Loll PJ, Garavito RM. The X-ray crystal structure of the membrane protein prostaglandin H2 synthase-1. Nature 1994;**367**:243-249

[56] Andronesi OC, Becker S, Seidel K, Heise H, Young HS, Baldus M. Determination of membrane protein structure and dynamics by magic-angle-spinning solid-state NMR spectroscopy. Journal of the American Chemical Society. 2005;**127**:12965-12974

[57] Cotton TM, Schultz SG, Van Duyne RP. Surface-enhanced resonance Raman scattering from cytochrome c and myoglobin adsorbed on a silver electrode. Journal of American Chemical Society. 1980;**102**:7960-7962

[58] Cotton TM, Van Duyne RP, Resonance Raman scattering from Rhodopseudomonas sphaeroides reaction centers absorbed on a silver electrode. FEBS Letter. 1982;**147**:81-84

[59] Feng M, Tachikawa H. Surface-enhanced resonance Raman spectroscopic characterization of the protein native structure. Journal of American Chemical Society. 2008;**130**:7443-7448

[60] Grabbe ES, Buck RP. Surface-enhanced Raman spectroscopic investigation of human immunoglobulin G adsorbed on a silver electrode. Journal of American Chemical Society.1989;**111**:8362-8366

[61] Combs ZA, Chang SH, Clark T, Singamaneni S, Anderson KD, Tsukruk VV. Label-free Raman mapping of surface distribution of protein A and IgG biomolecules. Langmuir. 2011;27:3198-3205

[62] Han XX, Jia HY, Wang YF, Lu ZC, Wang CX, Xu WQ, Zhao B, Ozaki Y. Analytical technique for label-free multi-protein detection based on western blot and surface-enhanced Raman scattering. Analytical Chemistry. 2008;**80**:2799-2804

[63] Han XX, Cai LJ, Guo J, Wang CX, Ruan WD, Han WY, Xu WQ, Zhao B, Ozaki Y. Fluorescein isothiocyanate linked immunoabsorbent assay based on surface-enhanced resonance Raman scattering. Analytical Chemistry. 2008;**80**:3020-3024.

[64] Han XX., Kitahama Y, Tanaka Y, Guo J, Xu W. Q, Zhao B, Ozaki Y. Simplified protocol for detection of protein−ligand interactions via surface-enhanced resonance Raman scattering and surface-enhanced fluorescence. Analytical Chemistry. 2008;**80**:6567-6572

[65] Han XX, Huang GG, Zhao B, Ozaki Y. Label-free highly sensitive detection of proteins in aqueous solutions using surface-enhanced Raman scattering. Analytical Chemistry. 2009;**81**:3329-3333

[66] Xu LJ, Zong C, Zheng XS, Hu P, Feng JM, Ren B. Label-free detection of native proteins by surface-enhanced Raman spectroscopy using iodide-modified nanoparticles. Analytical Chemistry. 2014;**86**:2238-2245

[67] Yu Y, Wang J, Lin J Q, Lin D, Chen WW, Feng SY, Huang ZF, Li YZ, Huang H, Shi H, Chen R. An optimized electroporation method for delivering nanoparticles into living cells for surface-enhanced Raman scattering imaging. Applied Physics Letters. 2016;**108**. DOI: 10.1063/1.4947009

[68] Wang J, Zeng YY, Lin JQ, Lin L, Wang XC, Chen GN, Huang ZF, Li BH, Zeng HS, Chen R. SERS spectroscopy and multivariate analysis of globulin in human blood. Laser Physics. 2014;**24**:6

[69] Wang J, Lin D, Lin JQ, Yu Y, Huang ZF, Chen YP, Lin JY, Feng SY, Li BH, Liu NR, Chen R. Label-free detection of serum proteins using surface-enhanced Raman spectroscopy for colorectal cancer screening. Journal of Biomedical Optics. 2014;**19**:087003

[70] Wang YL, Vaidyanathan R, Shiddiky MJA, Trau M. Enabling rapid and specific surface-enhanced Raman scattering immunoassay using nanoscaled surface shear forces. ACS Nano. 2015;**9**:6354-6362

[71] Reza KK, Wang J, Vaidyanathan R, Dey S, Wang YL, Trau M. Electrohydrodynamic-induced SERS immunoassay for extensive multiplexed biomarker sensing. Small. 2016. DOI: 10.1002/smll.201602902

[72] Wang YL, Trau M. Accurate and sensitive total genomic DNA methylation analysis from sub-nanogram input with embedded SERS nanotags. Chemical Communication. 2016;**52**:3560-3563

[73] Wang YL, Wee EJ, Trau M. Highly sensitive DNA methylation analysis at CpG resolution by surface-enhanced Raman scattering via ligase chain reaction. Chemical Communication. 2015;**51**:10953-10956

[74] Wang YL, Rauf S, Grewal YS, Spadafora LJ, Shiddiky MJA, Cangelosi GA, Schlucker S, Trau M. Duplex microfluidic SERS detection of pathogen antigens with nanoyeast single-chain variable fragments. Analytical Chemistry. 2014;**86**:9930-9938

[75] Wang YL, Salehi M, Schutz M, Schlucker S. Femtogram detection of cytokines in a direct dot-blot assay using SERS microspectroscopy and hydrophilically stabilized Au-Ag nanoshells. Chemical Communication. 2014;**50**:2711-2714

[76] Wang YL, Salehi M, Schutz M, Rudi K, Schlucker S. Microspectroscopic SERS detection of interleukin-6 with rationally designed gold/silver nanoshells. Analyst. 2013; **138**:1764-1771

[77] Ni J, Lipert RJ, Dawson GB, Porter MD. Immunoassay readout method using extrinsic Raman labels adsorbed on immunogold colloids. Analytical Chemistry. 1999;**71**:4903-4908

[78] Driskell JD, Kwarta KM, Lipert RJ, Porter MD, Neill JD, Ridpath JF. Low-level detection of viral pathogens by a surface-enhanced Raman scattering based Immunoassay. Analytical Chemistry. 2005;**77**:6147-6154

[79] Grubisha DS, Lipert RJ, Park HY, Driskell J, Porter MD. Femtomolar detection of prostate-specific antigen: An immunoassay based on surface-enhanced Raman scattering and immunogold labels. Analytical Chemistry. 2003;**75**:5936-5943

[80] Driskell JD, Uhlenkamp JM, Lipert RJ, Porter MD. Surface-enhanced Raman scattering immunoassays using a rotated capture substrate. Analytical Chemistry. 2007; **79**:4141-4148

[81] Wang G, Lipert RJ, Jain M, Kaur S, Chakraboty S, Torres MP, Batra SK, Brand RE, Porter MD. Detection of the potential pancreatic cancer marker MUC4 in serum using surface-enhanced Raman scattering. Analytical Chemistry. 2011;**83**:2554-2561

[82] Wang G, Park HY, Lipert RJ, Porter MD. Mixed monolayers on gold nanoparticle labels for multiplexed surface-enhanced Raman scattering based immunoassays. Analytical Chemistry. 2009;**81**:9643-9650

[83] Cao YC, Jin R, Nam JM, Thaxton CS, Mirkin CA. Raman dye-labeled nanoparticle probes for proteins. Journal of American Chemical Society. 2003;**125**:14676-14677

[84] Penn MA, Drake DM, Driskell JD. Accelerated surface enhanced Raman spectroscopy (SERS)-based immunoassay on a gold-plated membrane. Analytical Chemistry. 2013;**85**:8609-8617

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