

Review



Gold-Based SERS Platform for Biomedicine: Material Design, Enhancement Mechanisms, and Diagnostic Applications

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How To Cite: Zheng, S.; Cai, X.; Sun, L.; et al. Gold-Based SERS Platform for Biomedicine: Material Design, Enhancement Mechanisms, and Diagnostic Applications. *Nano-electrochemistry & Nano-photochemistry* **2026**, *2*(2), 13. <https://doi.org/10.53941/nenp.2026.100013>

Received: 3 April 2026

Revised: 5 June 2026

Accepted: 5 June 2026

Published: 18 June 2026

Abstract: Surface-enhanced Raman scattering (SERS) has emerged as a powerful analytical technique for biomedical detection owing to its ultrahigh sensitivity, molecular fingerprinting capability, excellent photostability, and potential for multiplex analysis. Among various SERS-active materials, gold-based systems have attracted particular attention due to their superior chemical stability, biocompatibility, and versatile surface functionalization. In this review, we systematically summarize the design strategies and enhancement mechanisms of gold-based SERS substrates. Specifically, we discuss electromagnetically dominated systems, as well as emerging platforms involving chemical enhancement and electromagnetic-chemical (EM-CM) synergistic effects, highlighting the evolution from classical plasmonic enhancement toward interface-mediated mechanisms. Furthermore, we provide a comprehensive overview of the biomedical applications of gold-based SERS platforms, with a focus on the detection of major diseases, including cancer, cardiovascular diseases, and neurological disorders. Finally, we critically analyze the key challenges hindering their practical implementation, such as reproducibility, stability in complex biological environments, and clinical translatability, and outline future perspectives for the development of gold-based SERS toward reliable and task-oriented biomedical applications.

Keywords: surface-enhanced Raman scattering; gold-based nanomaterials; material design; enhancement mechanism; biomedical diagnosis

1. Introduction

1.1. Fundamentals and Mechanisms of SERS

Raman spectroscopy is a spectroscopic analytical technique based on molecular vibrational information. It provides structural information through the detection of the inelastic scattering of incident light by molecules. When monochromatic light interacts with molecules, most photons undergo elastic scattering, namely Rayleigh scattering, while only a very small fraction interacts with the molecular vibrational or rotational energy levels, generating frequency-shifted scattering signals. This frequency shift constitutes the Raman signal [1]. Because



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different molecules possess distinct vibrational signatures, Raman spectroscopy can provide highly specific structural information and is therefore often referred to as a molecular “fingerprint” spectrum [2].

Raman spectroscopy has several unique advantages. First, it can provide highly specific molecular information. Second, because the Raman scattering of water molecules is weak, this technique is suitable for the analysis of samples in aqueous environments [3]. In addition, Raman detection usually does not require complicated pretreatment and causes relatively little damage to the sample. However, conventional Raman signals are inherently weak. The Raman scattering cross-section is extremely small, and only a tiny fraction of incident photons contributes to detectable Raman signals. This physical characteristic largely limits the direct application of conventional Raman spectroscopy in trace analysis and highly sensitive detection [4,5].

The emergence of surface-enhanced Raman scattering (SERS) significantly changed this situation. In 1974, Fleischmann et al. observed an abnormally enhanced Raman signal of pyridine molecules on a roughened silver electrode surface [6]. Subsequent studies showed that this enhancement did not simply arise from increased molecular adsorption, but rather from the amplification effect of the metal surface on the Raman scattering process. Since then, SERS has evolved into a highly sensitive molecular detection technique.

A large number of studies have shown that the enhancement factor (EF) of SERS can typically reach the order of 10^6 to 10^{14} , and under specific conditions, it can even enable single-molecule detection [7]. Such remarkable enhancement mainly arises from two mechanisms: electromagnetic enhancement (EM) and chemical enhancement (CM). EM originates from the localized surface plasmon resonance (LSPR) of metal nanostructures, which generates strongly enhanced local electromagnetic fields. Molecules located at plasmonic “hot spots”, such as interparticle gaps, tips, or rough surfaces, therefore exhibit greatly amplified Raman signals [8]. In contrast, CM results from molecule–metal interactions, including charge transfer and adsorption-induced changes in electronic structure and polarizability. Although CM generally contributes less than EM, it remains important in certain systems [9]. In practical SERS platforms, EM and CM often coexist and collectively determine the overall enhancement performance.

In the field of molecular detection, fluorescence-based methods have been widely used as conventional analytical approaches. Compared with traditional fluorescence detection, SERS also exhibits superior photostability and greater potential for multiplex detection. Fluorescent molecules are prone to photobleaching under continuous excitation, whereas SERS signals reflect molecular vibrational scattering processes with extremely short lifetimes and rapid relaxation, and thus generally show good photostability and are therefore more suitable for repeated measurements and continuous analysis [10,11]. Meanwhile, Raman spectral peaks are usually narrower and sharper than fluorescence emission bands, so different Raman reporter molecules can be clearly distinguished based on their characteristic peaks. This feature makes SERS inherently suitable for multiplex detection [12].

Despite these advantages, the development of SERS technology still faces several key challenges. The enhancement capability varies greatly among different substrates, the reproducibility of signals still needs to be improved, and stable detection in complex environments remains a major challenge [7]. Therefore, the construction of SERS substrates with high EF, high stability, and good reproducibility has always been a central issue of sustained interest in this field.

1.2. SERS for Biomedical Diagnosis: Opportunities and Challenges

In recent years, SERS has attracted extensive attention across a wide range of analytical fields, including environmental monitoring, food safety, chemical sensing, and bioanalysis. Among these, biomedical diagnosis is of particular importance due to its close relevance to human health. Early diagnosis of major human diseases has always been a central focus in medical research. High-burden diseases such as cancer, neurological disorders, and cardiovascular diseases have long posed serious threats to global health [13–15]. Many of these diseases often lack obvious symptoms at early stages, and patients are frequently diagnosed only after the disease has progressed to the middle or late stages, which increases treatment difficulty and significantly worsens the prognosis. Therefore, the medical community has consistently emphasized the importance of “early detection, early diagnosis, and early intervention” [16].

The realization of early diagnosis depends on highly sensitive analytical techniques. During the occurrence and progression of many diseases, the associated biomarkers often exhibit changes only at extremely low concentrations [17–19]. These biomarkers may be present in blood, urine, cerebrospinal fluid, or other body fluids, and their concentrations are often at the picomolar or even femtomolar level [20,21]. At the same time, nonspecific adsorption, background interference, and multicomponent interference in complex samples can further affect the detection accuracy. Therefore, the development of detection techniques capable of direct, rapid, and sensitive

identification of trace molecules has become an important direction at the interface between analytical science and clinical medicine [22–24].

Against this background, SERS has emerged as one of the most promising technologies in biomedical detection because it combines high sensitivity, molecular fingerprinting capability, good photostability, and multiplex detection potential [25]. SERS technology has been widely applied in the highly sensitive detection and phenotypic analysis of biomarkers associated with major diseases. In some studies, SERS platforms have even enabled reliable detection at the femtomolar level and below, demonstrating their potential for early disease screening and precision analysis [26–28].

However, for SERS to truly serve medical diagnosis, several key issues still need to be addressed. For example, because biological samples are highly complex, the stability and anti-interference capability of substrates in practical systems still need to be improved; the reproducibility and quantitative capability among different platforms also require further optimization. Therefore, constructing SERS substrates with high enhancement capability, reliable stability, good reproducibility, and compatibility with real biological samples remains crucial for promoting the medical application of this technology [10,29,30].

1.3. Au-Based Materials for SERS Platform

Among various strategies developed to improve SERS performance, the design of substrate materials plays a pivotal role. Noble metals are generally regarded as the most classical SERS substrate materials. These materials possess a high density of free electrons, which enables the excitation of surface plasmon resonance under incident light and the generation of significantly enhanced localized electromagnetic fields on the nanostructure surface. This characteristic provides an important basis for Raman signal amplification. Therefore, noble metals such as gold (Au) and silver (Ag) have long occupied a central position in SERS substrate research [7,9,31].

Among common noble metals, Au exhibits favorable properties that are well-suited for biomedical detection [32]. Au nanostructures usually show significant optical responses in the visible to near-infrared region. By regulating their size, morphology, and local structure, researchers can alter their absorption and scattering characteristics and further tune the distribution of localized electric fields [33–35]. Compared with silver, Au generally has higher chemical stability in air and aqueous environments, and its surface is less prone to oxidation. This feature makes Au-based materials more suitable for long-term storage, detection in complex media, and bioanalysis [36].

In addition to stability, Au materials also possess good biocompatibility and surface modifiability [37]. Meanwhile, the Au surface can readily bind with functional groups such as thiol, amino, and carboxyl groups [38]. Molecules such as antibodies, aptamers, peptides, and polymers can also be stably modified onto their surfaces [39,40]. It is these favorable interfacial chemical properties that make Au an important material platform for constructing functionalized SERS probes.

Au nanostructures also exhibit good synthetic controllability [41]. Researchers can prepare a variety of structures, such as nanospheres, nanorods, nanostars, and nanoflowers, by using wet-chemical methods [42]. Different morphologies present different plasmonic responses and hotspot distribution characteristics [43]. This provides a rich material basis for tuning enhancement effects and expanding application scenarios. Therefore, Au has become one of the most commonly used SERS substrate materials in the field of biomedical detection.

This review focuses on the structural design, enhancement mechanisms, and application progress of Au-based SERS substrates in biomedical detection. Section 2 mainly introduces Au-based SERS substrates dominated by EM, with emphasis on the research basis and structural classification of pure Au nanostructures and noble metal composite structures. Section 3 further discusses Au-based composite systems involving CM effects, as well as the development of Au clusters, single atoms, and other emerging Au-based SERS materials. Section 4 systematically summarizes the research progress of Au-based SERS substrates in the detection of major diseases, including cancer, neurological disorders, and cardiovascular diseases, from the perspective of disease-oriented applications. Finally, Chapter 5 outlines the major challenges currently faced by Au-based SERS substrates in practical applications and provides an outlook on their future development directions.

2. Electromagnetic Enhancement Dominated Au-Based SERS Systems

2.1. Design Principles and Structural Classification

Among Au-based SERS material systems, substrates dominated by EM are the earliest developed and the most representative class. The enhancement effect of such systems mainly originates from the LSPR induced by nanostructures and the resulting hotspot electric fields, which directly correlate SERS performance with structural

parameters [44]. These substrates are of fundamental research value for understanding the relationship between hotspot formation, signal amplification, and structural engineering in Au-based SERS materials [45].

The main research line of this type of Au-based SERS substrate has primarily focused on the construction and regulation of localized electromagnetic fields. Researchers usually optimize localized surface plasmon resonance behavior by tuning particle morphology, interparticle spacing, surface roughness, and assembly configuration, thereby further increasing the number, intensity, and accessibility of hotspots [46]. With the continuous deepening of research, the design goals of these substrates have gradually expanded from simply pursuing high enhancement effects toward also emphasizing biocompatibility, sample adaptability, and detection reliability [29,47]. This shift indicates that Au substrates dominated by EM are no longer merely basic materials for signal amplification, evolving into functional platforms optimized for task-specific detection [26].

From the structural perspective, pure Au SERS substrates dominated by electromagnetic enhancement can mainly be divided into two categories: colloidal systems and solid systems [44]. Colloidal Au substrates are usually composed of dispersed gold nanoparticles (AuNPs), particles with special morphologies, and their aggregates. These systems offer flexible surface functionalization and can be readily combined with antibodies, aptamers, nucleic acids, or other recognition units. At the same time, colloidal particles exhibit good dispersibility and fluidity, facilitating their interaction with target analytes in liquid-phase or complex biological environments, and thus making them suitable for in situ recognition and dynamic monitoring. In contrast, solid gold substrates rely on the immobilized construction of nanostructures at interfaces [48]. Such systems are more likely to provide stable detection interfaces and uniform signal outputs, offering distinct advantages in reproducible analysis, complex sample detection, and standardized applications.

In addition to the two types of substrates mentioned above, core-shell Au-based SERS tags are also an important structural form worthy of separate consideration [49], although some of them may also exist as colloidal nanoparticles in physical form. This separation is based on their distinct structural and analytical functions rather than only on their dispersion state. In core-shell systems, the core-shell interface, shell thickness, shell composition, surface roughness, and multilayer architecture can be deliberately engineered to regulate plasmon coupling, hotspot distribution, reporter molecule embedding, and tag stability. Therefore, these systems are particularly important for constructing bright and stable SERS labels for immunoassays, multiplex detection, and bioimaging [50,51].

Although these substrate systems differ in construction strategies and application scenarios, their core design concept remains consistent: to regulate hotspots through structural engineering and thereby enhance analytical performance in real detection settings. In the following, we discuss the research progress of Au-based SERS materials dominated by electromagnetic enhancement across colloidal substrates, solid substrates, and core-shell architectures, as well as emerging optical fiber-based SERS platforms, with an emphasis on design principles, structural features, and biomedical applications.

It should be noted that the “Au-based SERS substrates dominated by EM” discussed in this chapter mainly refer to SERS systems in which Au serves as the only or primary plasmonically active component, and whose enhancement behavior remains primarily dominated by localized electromagnetic field amplification. Although some studies introduce auxiliary supporting structures such as polymers, hydrogels, paper-based materials, flexible substrates, or microfluidic chips, these components are mainly used for interface construction, sample transport, signal regulation, or mechanical support and thus fall within the scope of this discussion [36,52].

2.2. Colloidal Au-Based SERS Systems

Regular spherical AuNPs are often used as basic models for studying the enhancement behavior of gold-based colloidal SERS systems because of their simple structure and relatively mature synthesis methods. However, such structures have limited variation in surface curvature, and the number of hotspots as well as the field enhancement capability are usually constrained to some extent. Therefore, this section will not further discuss regular spherical substrates, but will instead focus on pure gold colloidal substrates with complex surface morphologies. Pure gold colloidal substrates with complex surface morphologies can generate hotspots on the particle surface through spikes, branches, protrusions, or high-curvature structures, thereby producing stronger localized electromagnetic fields. As a result, they often exhibit higher optical response activity than smooth or regularly shaped pure gold particles [31,53,54].

Within this framework, a variety of probes with high hotspot density and diverse morphologies have been developed [53]. Among them, AuNPs with spiky surfaces are relatively common [55,56]. For example, urchin-like AuNPs, whose surfaces are densely covered with spike structures, are favorable for hotspot formation and are particularly suitable for constructing liquid-phase immunodetection systems. For instance, Khosroshahi et al.

developed an oriented antibody-conjugated gold nanourchin colloidal SERS immunoprobe, using gold nanourchins with spike-rich surfaces as the plasmonic enhancement core, and achieving the oriented immobilization of anti-CA15-3 monoclonal antibodies on the nanoparticle surface through polyethylene glycol (PEG)/ADH modification [57]. Compared with random conjugation, this strategy improved the accessibility of antigen-binding sites and thereby produced a stronger SERS response. The authors applied this probe to the detection of CA15-3 in the serum of breast cancer patients and further investigated the effect of laser polarization on the enhanced signal.

A similar example is the gold nanostar. Li et al. constructed a DNAzyme-functionalized gold nanostar SERS-fluorescence dual-modal nanoprobe for the detection and imaging of Ca^{2+} in living cells. In this system, the gold nanostar simultaneously served as both the SERS enhancement core and the fluorescence quenching platform [58]. Upon activation of the DNAzyme by Ca^{2+} , the Cy5-labeled substrate strand was cleaved and subsequently moved away from the particle surface, resulting in a decrease in the SERS signal and a recovery of fluorescence. The study showed that the detection limit of this probe was $0.056 \mu\text{M}$ in the fluorescence mode and as low as $0.021 \mu\text{M}$ in the SERS mode, and it could be used to monitor changes in intracellular Ca^{2+} levels during T-2 toxin-induced apoptosis. Similarly, Xiang et al. developed a gold nanostar-based liver-targeting SERS probe (GLTTs) for the *in vivo* diagnosis of early-stage liver fibrosis in mice (Figure 1A) [59]. In this system, gold nanostars served as the SERS-enhancing core, and glycyrrhetic acid (GA)-PEG-SH was further modified onto their surface through Au-S bonds, endowing the probe with good biocompatibility, prolonged circulation time *in vivo*, and specific recognition toward hepatic parenchymal cells. This probe was able to distinguish normal liver tissue from S1-stage fibrotic liver tissue, and characteristic peaks at 912, 1041, and 1340 cm^{-1} were identified as potential markers for early liver fibrosis, reflecting metabolic changes in carbohydrates, lipids, and proteins.

Researchers have also developed a single-particle dark-field scattering biosensing platform based on the surface etching of helical gold nanorods (HG NRs) and applied it to the highly sensitive detection of hepatitis B virus DNA (HBV-DNA) [60]. In this system, HG NRs with protruded and helical surface structures were used as specially shaped pure gold probes. Through the hybridization chain reaction (HCR) reaction triggered by HBV-DNA and the $\cdot\text{OH}$ generated by G4-DNAzyme catalysis, the surface of the HG NRs was etched, thereby causing changes in single-particle color and scattering intensity. Compared with ordinary Au NRs, HG NRs exhibited better detection performance toward HBV-DNA because of their larger specific surface area, higher surface activity, and stronger localized field response, with a linear range of 0.05–10 pM and a detection limit as low as 30.15 fM, which was markedly superior to the 1115.51 fM achieved by Au NRs.

While the above studies mainly relied on spikes, branches, and high-curvature regions on single-particle surfaces to achieve enhancement, gold particle assemblies enable more designable hotspots by artificially constructing controlled gaps. Along this route, the research focus has shifted from “optimizing the surface of single particles” to “precisely regulating the location and size of interparticle hotspots,” thereby allowing the recognition process to better match the spatial distribution of hotspots [61]. Sharma et al. used DNA origami to precisely assemble two gold nanorods into a dimeric SERS nanoantenna with a nanoscale gap, and further introduced an epidermal growth factor receptor (EGFR) aptamer so that the target protein could be directionally captured into the plasmonic hotspot region (Figure 1B) [62]. The data reported in the article showed that this platform was able to obtain the intrinsic SERS signal of EGFR, with a detection limit of 0.2 nM, and exhibited good recognition selectivity in the presence of non-target proteins such as bovine serum albumin (BSA), myoglobin, and vascular endothelial growth factor (VEGF) in the analyte. This study indicates that gold particle assembly systems can achieve effective utilization of hotspot space through precise assembly and can effectively couple recognition units with the hotspot region, thereby improving the feasibility of label-free protein detection.

Some studies have also obtained detection information in a reverse manner by disrupting hotspots. Su et al. presented a nucleic acid-linked aggregated SERS system and combined it with the CRISPR/Cas12a molecular recognition mechanism to construct an AuNP aggregation-based SERS detection platform [63]. This system consisted of AuNP probes surface-modified with DNA and the Raman reporter molecule mercaptobenzonitrile (MBN), linker ssDNA, and a Cas12a/crRNA recognition module. In the absence of target DNA, intact linker ssDNA induced AuNP aggregation, thereby generating a strong SERS signal; when target double-stranded DNA was present, Cas12a was activated and cleaved the linker ssDNA, causing the probes to remain dispersed and resulting in a decreased SERS signal. Using HPV16 and HPV18 as models, the authors achieved detection at the pM level, with a total detection time of only about 40 min. Unlike the static hotspot platform assembled by DNA origami, this system placed greater emphasis on signal changes caused by transitions between aggregated and dispersed states, indicating that gold particle assemblies are also suitable for coupling with nucleic acid recognition and enzymatic reactions to form responsive SERS detection platforms.

Overall, although the above studies targeted different analytes, their core principles are consistently based on the regulation of hotspot engineering through the morphology or aggregation configuration of gold nanostructures. Gold particles with special morphologies place greater emphasis on enhancing the single-particle enhancement capability through spikes, branches, and surface curvature, and are further extended to various biological environments with the aid of functional units such as antibodies and ligands. In contrast, gold particle assemblies rely more on controllable gaps or responsive aggregation behavior to achieve hotspot location design and signal regulation. Taken together, these studies highlight that the development trend of gold-based colloidal SERS materials has shifted from the simple pursuit of high EF to a broader focus on hotspot controllability, surface recognition efficiency, adaptability to complex samples, and multifunctional integration, thereby forming an important basis for their continued advancement toward precise biomedical detection [64,65].

For colloidal Au-based SERS systems, their main advantages lie in flexible surface functionalization, efficient interaction with analytes in solution, and suitability for liquid-phase recognition scenarios, such as biomarker detection, nucleic acid sensing, exosome analysis, and cell targeting. However, these systems are also highly dependent on particle aggregation state, ionic strength, protein corona formation, nonspecific adsorption, and batch-to-batch variation. In many cases, extremely low detection limits are obtained under highly controlled laboratory conditions, whereas their quantitative reproducibility in real biological matrices remains insufficiently validated. Therefore, the future development of colloidal Au-based SERS probes should focus not only on increasing hotspot density, but also on improving colloidal stability, antifouling capability, standardized surface chemistry, and reproducible signal output in complex samples.

2.3. Solid Au-Based SERS Systems

In contrast to colloidal systems, the development of solid Au-based SERS substrates is more oriented toward the integrated optimization of structural controllability, signal uniformity, and applicability to complex samples [66]. Compared with colloidal systems, solid substrates are more suitable for providing stable detection interfaces and are more readily integrated into standardized analytical workflows. Therefore, they show clear advantages in environmental analysis, complex matrix screening, and biomedical detection [67,68]. Currently, the development strategies of related substrates mainly include increasing hotspot density through structural engineering, improving reproducibility through array construction, and addressing the enrichment and recognition of target molecules in complex samples through functionalization strategies [66,69,70].

At the most fundamental level of material design, some studies have focused on how to construct high-density hotspots through the regulation of the combined structures of solid-state AuNPs. Pal et al. prepared a gold nanoisland substrate using a repeated dewetting strategy. Through a two-step 5 nm gold film deposition-annealing process, they obtained nanoisland structures with smaller sizes and narrower gaps, thereby significantly increasing the hotspot density [71]. Compared with a single-step dewetted substrate of equivalent thickness, this structure showed a markedly enhanced ability for Rhodamine 6G (R6G) detection, with a detection limit as low as 0.1 fM and an experimental EF reaching 4.61×10^{11} .

In addition, researchers have constructed a three-dimensional porous dendritic gold film/flexible indium tin oxide (ITO) composite substrate based on a three-dimensional structural design (Figure 1C) [72]. This substrate induced the self-assembly of AuNPs through deep eutectic solvent (DES)-assisted thermal evaporation, forming a dendritic porous Au film with high hotspot density, an open porous structure, and mechanical flexibility. The data showed that this system could not only achieve detection of crystal violet (CV) at concentrations as low as 6.4×10^{-15} M, but also enable sensitive analysis of poly(ethylene terephthalate (PET) and polystyrene (PS) nanoplastics, with detection limits of 0.051 $\mu\text{g/mL}$ and 8.2 $\mu\text{g/mL}$, respectively, and was further applicable to spiked sample analysis in complex samples such as tap water, lake water, diluted milk, and wine. Despite differences in structural form, both studies indicate that solid gold substrates can improve hotspot density and increase the probability of target molecules contacting hotspots through rational nanomorphology design, thereby achieving highly sensitive detection [73].

On the basis of developing gold-based solid SERS substrates with strong enhancement effects, some researchers have attempted to improve the uniformity of signal output and the capability for quantitative analysis through ordered arrays. Mo et al. constructed a gold micro/nano ordered array solid SERS substrate. By combining polystyrene monolayer template-assisted etching with magnetron sputtering, they obtained a long-range ordered structure composed of rough Au particles of about 100 nm, with an interparticle spacing of approximately 16.8 nm, thereby generating abundant and uniformly distributed hotspots [74]. The substrate exhibited an EF of up to 3.26×10^7 for 4-mercaptobenzoic acid (4-MBA), showed a good linear response to etomidate in e-cigarette oil over the range of 1–50 ppm, achieved a detection limit of about 17 ppb, and showed a relative standard deviation

(RSD) of only 2.83% for the 1003 cm^{-1} peak on the same substrate. The authors further demonstrated that this system could achieve rapid identification of etomidate in complex e-cigarette oil matrices and distinguish its metabolite, etomidate acid. This work indicates that ordered gold arrays can not only improve signal intensity, but also provide a stable and reproducible structural basis for rapid screening in complex samples.

Similarly, Lu et al. constructed a solid gold nanocone array substrate and applied it to serum fingerprint detection and disease diagnosis. This substrate was fabricated by polystyrene colloidal sphere template-assisted reactive ion etching and magnetron sputtering, featuring a uniform and controllable array structure as well as abundant inter-nanogap hotspots (Figure 1D) [75]. Notably, the authors further pointed out that the thickness and viscosity of the serum liquid layer could significantly affect the SERS stability on solid substrates. Therefore, they systematically optimized the serum dilution conditions and finally determined that a 50-fold dilution could provide the most complete serum spectra with better reproducibility. Under this condition, the substrate achieved an EF of 1.9×10^6 for R6G, with the RSD of the 490 cm^{-1} peak in serum detection being lower than 3.74%. The serum SERS spectra of healthy individuals could be distinguished from those of patients with gastric cancer or colorectal cancer, demonstrating stable and reproducible diagnostic performance in real biofluid analysis.

Beyond hotspot construction and array uniformity, gold-based solid SERS substrates often face more specific challenges in practical applications, such as signal fluctuation, insufficient stability in quantitative analysis, and the difficulty of allowing large molecular targets to fully enter hotspot regions. To address these issues, researchers have introduced strategies such as internal standard correction, interfacial regulation, and active delivery to further improve the reliability and sensitivity of solid platforms in specific analytical tasks. Tan et al. prepared an ordered array of spindle-shaped AuNPs through liquid-liquid interfacial self-assembly, immobilized carboxy-X-rhodamine (ROX)-labeled hairpin DNA H1 on its surface, and introduced 5,5'-dithiobis(2-nitrobenzoic acid) (DTNB) as an internal standard molecule to construct a dual-signal solid SERS biosensor for miRNA-21 detection [76]. When miR-21 was present, H1 was opened and caused ROX to move away from the gold surface, resulting in a decrease in the ROX signal, while the DTNB signal remained stable, thereby enabling self-calibrated detection. This system showed a good linear relationship in the range of 0.1 pM– 10^5 pM, with a detection limit as low as 0.046 pM, and the RSD of only 3.35% across 30 random sites. These results indicate that when the uniform hotspots provided by ordered arrays are combined with an internal standard correction strategy, the quantitative accuracy and reproducibility of solid SERS platforms can be further improved.

Gao et al. focused on another representative limitation, namely that macromolecules such as proteins cannot always efficiently enter the strongest hotspot regions on solid substrates. To address this issue, they constructed a gold nanotriangle array substrate based on thermoresponsive poly(N,N-diethylacrylamide) (pDEAA) hydrogel, in which hexagonally close-packed gold nanotriangles were transferred onto the hydrogel surface, and the interparticle gaps could be reversibly switched between “open” and “closed” states through temperature regulation (Figure 1E) [77]. On this basis, the authors further proposed a Gel Filter Trapping (GFT) strategy, which used the local water flow and sieving effect generated by hydrogel water absorption to actively deliver proteins into the nanogap hotspot regions. The study showed that this system enabled the detection of hemoglobin at concentrations as low as 100 fg/mL and provided a quantitative range spanning six orders of magnitude, while also showing good detection capability for myoglobin and lysozyme. By using a tunable gap structure, this study effectively improved the accessibility of proteins to SERS hotspots, and therefore provides a meaningful reference for the application of solid substrates in biomacromolecule detection.

Although these studies address different challenges, both indicate that the performance improvement of solid SERS substrates is not limited to the optimization of the nanostructure itself. When facing specific detection tasks, it is also necessary to combine corresponding strategies to solve key issues such as signal stability or target molecule accessibility, so that the application potential of solid substrates in analyte analysis can be more fully realized.

Overall, these studies collectively indicate that the core value of gold-based solid SERS substrates lies in constructing stable interfaces with high hotspot density through controllable solid-state nanostructures, and on this basis, improving analytical performance in complex samples. Although the related works differ in structural form, detection targets, and implementation strategies, their concerns are consistent, namely how to balance signal reproducibility, sample adaptability, and practical detection reliability while maintaining enhancement effects. Gold-based solid SERS substrates are not merely carriers for signal enhancement, but have evolved into functional analytical platforms that can integrate interfacial regulation, signal calibration, and task-oriented design, which lays the foundation for their further expansion in practical detection and biomedical applications [69,78].

Compared with colloidal systems, solid Au-based SERS substrates are more suitable for standardized spectral acquisition and reproducible analysis, especially when ordered arrays or lithographically defined structures are used. This makes them attractive for serum fingerprinting, biofluid discrimination, and device-integrated sensing. However, their superiority in reproducibility does not automatically guarantee biomedical applicability. In

practical assays, solid substrates may suffer from limited analyte-hotspot contact, uneven drying of biological fluids, surface fouling, and reduced accessibility for large biomolecules such as proteins or extracellular vesicles. Moreover, fabrication strategies that provide excellent uniformity at the laboratory scale may be costly or difficult to scale up. Therefore, the translational value of solid Au substrates depends on whether high signal uniformity can be combined with simple fabrication, efficient sample delivery, antifouling interfaces, and robust performance in real biological fluids.

2.4. Core-Shell Au-Based SERS Systems

Among gold-based SERS materials, core-shell structures also represent a typical enhancement architecture. Such systems usually use Au or Ag as the core component, and further regulate localized plasmon coupling and hotspot distribution by introducing another metallic shell, a rough outer surface, or multiple interfacial structures, thereby achieving higher SERS activity than individual AuNPs [49,79,80]. For gold-based SERS substrates intended for biodetection, the value of core-shell structures lies not only in enhancing signal intensity, but also in providing more stable embedded sites for Raman reporter molecules and improving the applicability of the tags in immunoassays and multiplex detection through interfacial design [81–83].

Some of these designs mainly focus on single-layer core-shell structures. Huang et al. developed a 4-MBA-labeled Ag@Au core-shell porous nanocage SERS tag and applied it to the highly sensitive sandwich immunodetection of alpha-fetoprotein (AFP) (Figure 1F) [84]. In this system, silver nanoparticles were used as sacrificial templates, and a rough, porous gold shell was formed through an in situ replacement reaction while part of the silver core was retained, thereby constructing a core-shell SERS tag that combined hotspots from the rough Au shell with the enhancement capability of the residual Ag core. The study showed that the SERS signal intensity of this Ag@Au porous nanocage was more than 10 times higher than that of conventional Au nanoparticle tags. In AFP immunoassays, it exhibited a detection range of 0.2–500 ng mL⁻¹ and a detection limit as low as 0.12 ng mL⁻¹. These results suggest that constructing more effective hotspots through a porous and rough shell structure is a direct and effective way to improve the detection performance of gold-based SERS substrates.

Similarly, Yang et al. developed an enzyme-induced Au@Ag core-shell nanostructure-enhanced SERS immunoassay system and applied it to the ultrasensitive detection of alpha-fetoprotein. In this method, 4-MBA-labeled AuNPs were first used as seeds, and then alkaline phosphatase was introduced through the immunoreaction catalyzed alkaline phosphatase (AAP) to generate vitamin C, which reduced Ag⁺ and deposited it onto the AuNP surface, thereby forming an Au@Ag core-shell structure with stronger SERS activity in situ [85]. This system exhibited a good linear response toward AFP in the range of 0.5–100 pg mL⁻¹, with a detection limit as low as 0.081 pg mL⁻¹. Moreover, the detection results for serum samples from liver cancer patients were consistent with those obtained by clinical chemiluminescence immunoassay, demonstrating the application potential of this type of SERS probe.

Building on single-layer core-shell structures, researchers have also begun to introduce multilayer interfaces and multiple shell designs in order to achieve higher brightness and better signal protection. Ning et al. constructed a highly active SERS tag system based on gold-silver-silver core-shell-shell nanosea cucumbers (GSSNTs) and applied it to the sensitive simultaneous detection of exosomes associated with multiple cancers [86]. In this tag, a gold nanosea cucumber (GNT) core was first formed by growing rough gold nanoprotusions on the surface of gold nanorods, followed by the introduction of a double silver shell. Two layers of Raman reporter molecules were then embedded at the core-shell and shell-shell interfaces, respectively, thereby constructing a multilayer core-shell tag that combined hotspots from the rough gold core, the enhancement effect of the silver shells, and interfacial signal protection. The study showed that the final GSSNTs exhibited signal intensities about 2.3 times higher than gold-silver nanosea cucumbers (GSNTs), 39.3 times higher than GNTs, and 151.7 times higher than GNRs. Based on this design, the aptamer-based SERS sensing system enabled the detection of exosomes derived from LNCaP, SKBR3, and HepG2, with detection limits as low as 26, 72, and 35 particles μL⁻¹, respectively.

Collectively, these studies demonstrate the structural advantages of gold-based core-shell composite tags from different perspectives. Multilayer interfaces, rough core-shell morphologies, and reporter molecule embedding strategies can significantly improve tag brightness, stability, and multiplex detection capability. Core-shell structures are not only effective means of enhancing localized electromagnetic fields and protecting signal molecules in gold-based SERS tags, but also provide an important material foundation for constructing highly sensitive bioanalytical platforms suitable for multi-target detection and complex samples.

From an application perspective, core-shell Au-based SERS tags are among the most promising structures for immunoassays, multiplexed biomarker detection, and bioimaging because Raman reporters can be embedded or protected within engineered interfaces. This design improves signal stability and reduces reporter leakage

compared with simple surface-adsorbed probes. However, the increased structural complexity also introduces new challenges. The reproducibility of shell thickness, surface roughness, reporter loading, and antibody conjugation can strongly affect tag-to-tag signal uniformity and quantitative accuracy. In addition, multilayer or bimetallic core-shell structures often require multi-step synthesis, which may complicate large-scale production and quality control. Therefore, future studies should not only report tag brightness and detection limits, but also provide more systematic evaluation of batch consistency, reporter stability, nonspecific adsorption, and performance in real biological samples.

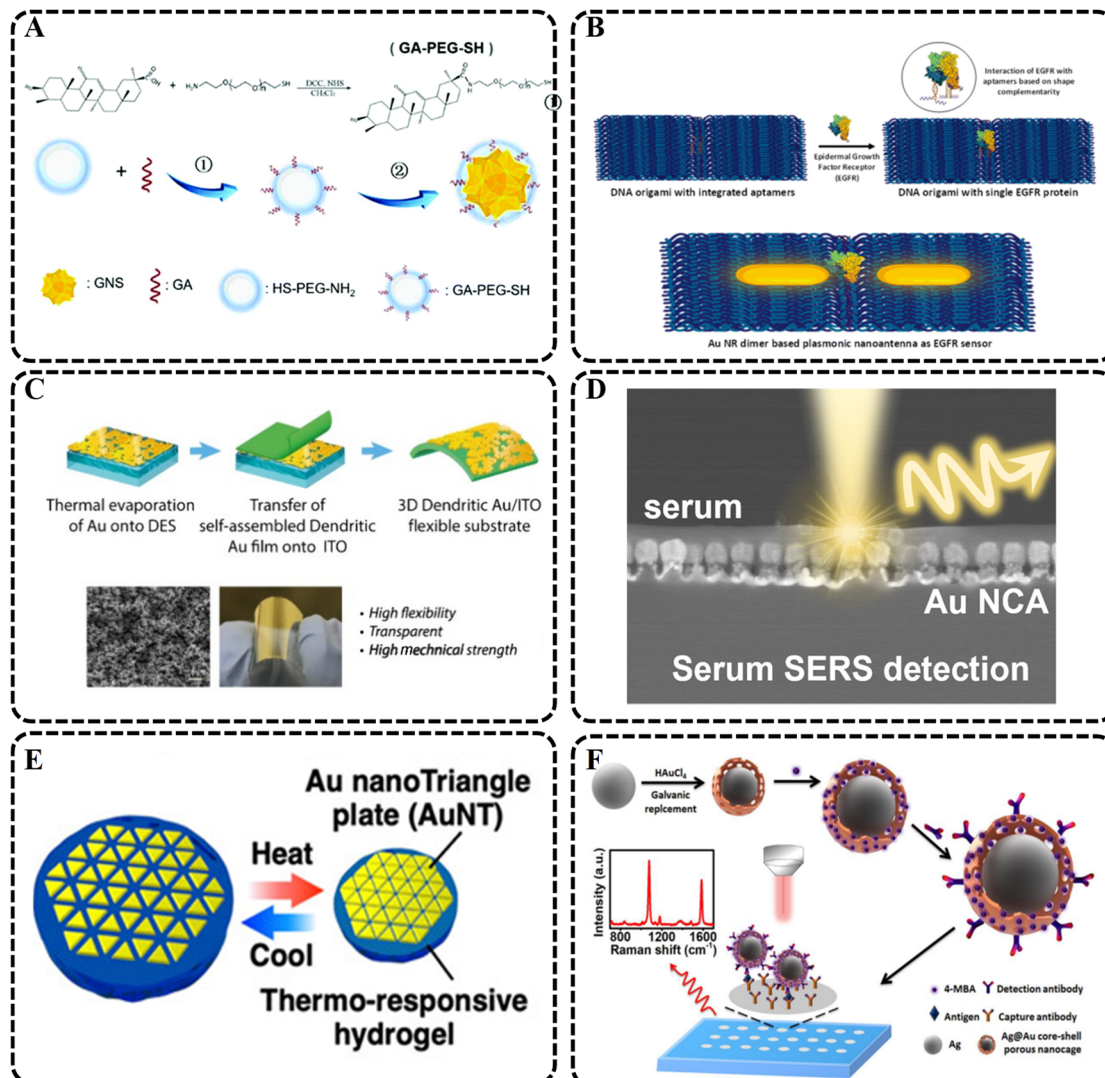


Figure 1. (A) Schematic illustration of a gold nanostar-based SERS nanoprobe for liver fibrosis detection. Adapted with permission from Ref. [58]. Copyright 2021, The Royal Society of Chemistry. (B) Gold nanorod dimer-based SERS nanoantenna assembled using a DNA origami template. Adapted with permission from Ref. [61]. Copyright 2024, The Royal Society of Chemistry. (C) Dendritic porous Au/ITO flexible SERS substrate fabricated via a deep eutectic solvent-assisted thermal evaporation and transfer strategy. Adapted with permission from Ref. [71]. Copyright 2024, American Chemical Society. (D) Au nanocone array substrate. Adapted with permission from Ref. [74]. Copyright 2023, American Chemical Society. (E) Thermo-responsive hydrogel-supported gold nanotriangle array SERS substrate for protein detection. Adapted with permission from Ref. [75]. Copyright 2024, American Chemical Society. (F) Construction of a 4-MBA-labeled Ag@Au core-shell porous nanocage SERS immunotag. Adapted with permission from Ref. [84]. Copyright 2019, by the authors. Licensee MDPI, Basel, Switzerland.

2.5. Au-Based Fiber Optic SERS Systems

In addition to colloidal probes, solid substrates, and core-shell SERS tags, Au-based optical fiber SERS optrodes and lab-on-fiber platforms represent an important class of SERS systems for biomedical sensing. In these systems, SERS-active nanostructures are integrated onto the distal end, side surface, inner wall, or microstructured region of an optical fiber, allowing excitation light delivery and Raman signal collection through the same or

coupled optical pathways [87]. Compared with conventional planar substrates, optical fiber SERS platforms provide several unique advantages, including miniaturization, remote signal acquisition, compatibility with narrow or confined spaces, and potential integration with endoscopic, catheter-based, or needle-based biomedical procedures [88]. Therefore, they are particularly attractive for minimally invasive sensing, in situ biochemical analysis, and possible in vivo measurements. Recent reviews on optical fiber biosensors and lab-on-fiber technology have also highlighted their potential in in vivo detection and precision biomedical diagnostics [89].

From the perspective of material design, Au nanostructures can be introduced onto optical fibers through several strategies, including the immobilization of Au nanoparticles or Au nanostars, sputtering or deposition of Au nanoislands, fabrication of ordered plasmonic nanostructures on the fiber tip, or growth of Au nanostructures inside hollow-core or microstructured fibers [90]. These designs transform the optical fiber from a passive light-guiding component into an active plasmonic sensing interface. For example, fiber-optrode SERS probes using silver-coated gold nanostars have been reported for highly sensitive molecular detection, showing how anisotropic Au-based nanostructures can be coupled with optical fibers to generate strong local electromagnetic fields at the sensing region. More recently, lab-on-fiber concepts have further emphasized the fabrication of engineered SERS optrodes, in which the fiber tip functions as a miniaturized sensing chip that integrates light delivery, plasmonic enhancement, and analyte interaction within a compact probe geometry [91].

The biomedical significance of optical fiber SERS platforms lies mainly in their application scenarios rather than in enhancement performance alone. Because optical fibers are flexible, compact, and compatible with remote optical interrogation, they can potentially be incorporated into endoscopic probes, catheters, fine-needle devices, or implantable sensing systems. This feature is particularly relevant for tissues or body sites that are difficult to access using conventional benchtop Raman microscopy. For example, SERS-assisted sandwich immunoassay platforms designed for integration with fine-needle aspiration biopsy have been reported for the detection of human thyroglobulin, indicating the potential of SERS optrode-like strategies for biomarker analysis in clinically relevant sampling workflows [92]. In addition, hollow-core microstructured optical fiber-based SERS systems have been explored for probing nanoscale biological targets such as exosomes, suggesting that fiber-integrated SERS platforms may provide a route toward highly sensitive liquid biopsy analysis in confined optical geometries [93].

Nevertheless, optical fiber SERS platforms also face specific challenges that differ from those of conventional colloidal or planar substrates. First, the reproducible fabrication of plasmonic nanostructures on a small fiber tip or within a microstructured fiber remains technically demanding, especially when uniform hotspot distribution and batch-to-batch consistency are required. Second, the small sensing area may limit analyte capture efficiency, making surface functionalization, local enrichment, and antifouling modification particularly important for complex biological samples. Third, for in vivo or endoscopic applications, issues such as probe sterilization, mechanical robustness, biocompatibility, background Raman signals from tissues or fibers, optical alignment, and quantitative calibration must be carefully considered. Therefore, although gold-based optical fiber SERS optrodes are highly promising for minimally invasive and in situ biomedical sensing, their translational value depends on whether stable nanofabrication, reliable surface chemistry, and clinically compatible operation protocols can be established.

3. EM–CM Synergistically Enhanced and CM-Dominated Au-Based SERS Systems

Beyond purely EM-dominated systems, an important direction is to integrate chemically active interfaces, such as semiconductors or carbon materials, to enhance interfacial interactions and complementary Raman contributions. Beyond purely EM-dominated systems, another important research direction is to extend the enhancement mechanism and interfacial functions of traditional gold substrates by integrating materials related to chemical enhancement.

3.1. Mechanistic Basis of EM-CM Synergistic Enhancement

In Au-based composite SERS systems, semiconductors or carbon components can complement EM enhancement by modulating molecular adsorption, interfacial charge transfer, and local electronic structure, resulting in EM-CM synergistic enhancement [94–97]. As a result, an important feature of such composite substrates is that their enhancement mechanism is typically governed by the synergistic effect of EM and CM [98].

In such systems, the synergistic effect can influence the stability of SERS signals and the detection sensitivity. The introduction of a second component can alter the distribution of target molecules on the substrate surface, making them more likely to approach hotspot regions or adsorb more stably on the composite interface. This not only helps increase the number of effectively detected molecules, but also helps reduce signal instability caused by fluctuations in adsorption states [95]. In certain cases, the second component can even endow the substrate with

additional functions such as photoresponse, self-cleaning, molecular sieving, or directional enrichment, thereby further improving the applicability of the platform in continuous detection or complex environments [99].

It should be noted that not all composite materials can automatically generate an ideal EM-CM synergistic enhancement effect [100]. The presence and extent of such synergy still depend on the electronic structure of the second component, the contact state between Au and that component, and the adsorption characteristics of the target molecules [94,101]. Only when the second component can truly participate in interfacial interactions and influence the Raman scattering process can the composite substrate evolve beyond a simple “combination of materials” to form a functional platform with synergistic enhancement significance [102]. Therefore, the design and regulation of composite interfaces have become key issues in the research of gold-based composite SERS materials [103–105].

Based on the above understanding, the following sections will focus on two types of gold-based systems related to CM. First, EM-CM synergistically enhanced composite substrates constructed from Au and semiconductors, carbon materials, or other components will be introduced. Subsequently, emerging gold-based substrates characterized mainly by CM, such as gold clusters and gold single atoms, will be discussed, in order to illustrate that as the size of gold is further reduced, the enhancement mechanism of gold-based SERS materials extends from “hotspot-dominated” to “interfacial electronic process-dominated.”

3.2. EM-CM Synergistically Enhanced Au-Based SERS Systems

3.2.1. Au-Semiconductor Composite Systems

Au–semiconductor composite SERS substrates represent one of the most typical systems exhibiting EM-CM synergistic enhancement [106,107]. In these materials, Au usually serves as the primary plasmonic enhancement unit, whereas semiconductors such as MoS₂, TiO₂ and ZnO can provide contributions from chemical enhancement and additional functions to the composite platform through interfacial charge transfer, band coupling, regulation of molecular adsorption, or surface modifiability [108–111].

From the perspective of interfacial design, some studies have mainly focused on how the combination of a single semiconductor with Au influences enhancement performance and interfacial electronic interactions. Yu et al. constructed a hierarchical rose-like MoS₂/Au nanocomposite SERS substrate by first preparing MoS₂ nanoflowers through a hydrothermal method and then growing Au nanoparticles in situ on their surface, thereby forming a composite platform that combines the charge-transfer characteristics of the semiconductor with the plasmonic enhancement effect of the metal (Figure 2A) [112]. It was found that the introduction of Au not only brought about a stronger localized surface plasmon resonance response, but also produced an n-type modulation effect on MoS₂, thereby improving the interfacial electronic structure and the molecule–substrate interaction. The study showed that the optimized MoS₂/Au-6 substrate achieved a detection limit of 2.1×10^{-9} M for CV, with an EF of about 8.52×10^6 , while also exhibiting good reproducibility.

Similarly, some researchers constructed an AuNPs/TiO₂ nanotube array composite substrate, in which TiO₂ nanotube arrays served as an ordered supporting framework, and Au nanoparticles were loaded onto the surface through gold sputtering and photoreduction, thereby forming a gold/semiconductor composite platform with both plasmonic enhancement capability and structural uniformity [113]. Furthermore, by taking advantage of the easily modifiable TiO₂ surface, the authors introduced octadecylphosphonic acid (ODPA) to convert the substrate into a hydrophobic interface, thereby promoting the enrichment of hydrophobic bacterial metabolites such as dimethyl disulfide (DMDS) in hotspot regions. The study showed that this composite substrate achieved a detection limit of 10^{-9} M for PA, with an EF of about 4.0×10^7 , and its SERS performance was superior to that of an AuNP-modified silicon substrate. Both of these studies indicate that the combination of gold with a single semiconductor can, to some extent, improve the detection interface and signal output of pure gold substrates.

In Au-semiconductor composite SERS substrates, ZnO represents another typical semiconductor material, and its integration with Au can significantly improve chemical enhancement. The n-type semiconductor properties and tunable band structure of ZnO enable synergistic interaction with Au nanostructures through interfacial charge transfer (CT), thereby strengthening molecule-substrate interactions and improving the intensity and stability of Raman signals. Liu et al. reported Au-ZnO composite nanoparticles as recyclable SERS substrates, which exhibited strong CT-induced enhancement and good reusability for the detection of R6G [110]. Yang et al. investigated the Au/ZnO/PATP system and found that ZnO made a significant contribution to interfacial charge transfer, effectively enhancing the SERS signal of PATP while also improving the adsorption mode of molecules at the composite interface [111]. In addition, previous reviews have indicated that, similar to TiO₂, ZnO can further enhance the SERS response and biological detection performance of Au-semiconductor composite substrates through nanomorphology regulation, photogenerated carrier transfer, and optimization of surface active sites [108].

Therefore, when designing Au-semiconductor composite SERS substrates, ZnO-based systems represent an important option for improving CM enhancement and reproducibility because of their unique electronic structure and chemical stability.

Beyond single semiconductor composites, other studies have shifted their focus to more complex hybrid semiconductor interfaces, aiming to further expand the sensing capability and application scope of heterojunctions through multi-interface synergy. Wei et al. developed a $\text{MoS}_2@\text{TiO}_2@\text{Au}$ heterojunction-based composite SERS platform and applied it to the detection of antifungal drug residues as well as the monitoring of photocatalytic degradation (Figure 2B) [114]. In this system, $\text{MoS}_2@\text{TiO}_2$ served as the semiconductor framework, onto which Au nanoparticles were further loaded, thereby integrating the charge-carrier separation advantage of the semiconductor components with the plasmonic enhancement effect of Au. The study showed that this platform enabled sensitive and reproducible SERS analysis of methylene blue (MB), CV, and malachite green (MG), and could also achieve relatively rapid self-cleaning under sunlight by utilizing the photocatalytic effect. When used for MB detection in shrimp protein extract, the detection limit reached $1.509 \mu\text{g/L}$. Compared with composite systems involving a single semiconductor, this type of heterojunction structure more clearly reflects researchers' exploration of interfacial electronic regulation and functional integration strategies.

Overall, Au-semiconductor composite systems provide a useful route to extend SERS enhancement beyond conventional electromagnetic hotspots. Through band alignment, interfacial charge transfer, photocatalytic activity, and surface adsorption regulation, semiconductors can improve molecular interaction with Au-based plasmonic structures and may introduce additional functions such as enrichment or self-cleaning. However, the interpretation of EM-CM synergy in these systems should be cautious. Changes in SERS intensity may arise not only from true charge-transfer enhancement, but also from altered molecular adsorption amount, surface area, local pH, or photochemical reactions. In addition, semiconductor components may introduce interfacial heterogeneity and possible photoinduced damage to biomolecules. Therefore, for biomedical applications, Au-semiconductor substrates should be evaluated not only by EF, but also by mechanistic evidence, stability under biological conditions, and compatibility with fragile biomolecules.

3.2.2. Au-Carbon Composite Systems

Beyond semiconductor-based systems, carbon materials have also attracted considerable attention. Notably, graphene and graphene oxide can promote the enrichment of target molecules at the interface through π - π interactions, electrostatic interactions, or their large specific surface area. Meanwhile, they can also provide fluorescence quenching, atomic-level isolation, and interfacial electronic coupling to a certain extent [115–118]. As a result, although such systems still rely on Au to provide the main plasmonic hotspots, the carbon materials act not merely as passive supporting layers, but directly influence the interaction mode between the composite substrate and the target molecules, as well as the final quality of the SERS readout.

From the viewpoint of interfacial engineering, some studies have focused on how the combination of gold with graphene/graphene oxide (GO) influences molecular adsorption and signal enhancement. Zhu et al. constructed a graphene-covered gold nanohole array composite SERS platform. This system combines the strong localized plasmonic field of the gold nanohole array with the atomically flat and chemically inert interface of graphene (Figure 2C) [119]. The study showed that graphene coverage could bring about a 30% increase in absorption and up to a 700-fold enhancement of the graphene Raman signal. At the same time, graphene could also serve as an atomic-scale isolation layer, separating molecules from the bare gold surface, thereby reducing spectral distortion caused by photocarbonization, photobleaching, and gold-surface catalysis. Furthermore, when R6G was placed on the graphene surface, the authors observed that, compared with the bare gold nanohole substrate, the composite substrate could provide cleaner and more reproducible molecular fingerprint signals. This work indicates that, compared with pure gold nanohole substrates, the combination of gold with graphene can not only enhance light-matter interactions, but also improve the stability of the detection interface and the fidelity of the signal.

Other studies have further taken advantage of the molecular enrichment capability of GO or graphene to construct gold-carbon composite platforms for real-sample analysis. Pan et al. developed a GO/gold nanostar hybrid paper-based SERS biosensor and applied it to the label-free detection of free bilirubin in serum (Figure 2D) [120]. This system combined the large specific surface area and π - π /electrostatic enrichment capability of GO with the high SERS activity of gold nanostars, thereby simultaneously achieving bilirubin enrichment, background suppression, and Raman signal amplification. Among them, the detection performance of the GO-gold nanostar (GO-GNS) composite was significantly better than that of GO alone or GNS alone.

spectra of R6G on various substrates. Adapted with permission from Ref. [119]. Copyright 2013, American Chemical Society. (D) Schematic of the operating principle for the enPSERS biosensor in label-free serum bilirubin detection, encompassing the preparation and sensing mechanism based on GO-GNS hybrids, structural and spectral analysis of bilirubin, and optical/SERS imaging characterization of the biosensor. Adapted with permission from Ref. [120]. Copyright 2019, Elsevier. (E) Comparison of SERS sensitivity and intensity between 3D crumpled and flat graphene-Au NP structures, encompassing SERS measurements of 4-MPH at varying concentrations, electromagnetic field enhancement simulations via COMSOL, and corresponding field enhancement line profile analyses. Adapted with permission from Ref. [121]. Copyright 2015, American Chemical Society.

Au-carbon composite systems are particularly valuable for label-free detection and complex-sample analysis because graphene, graphene oxide, and related carbon materials can enrich aromatic or biomolecular targets, quench fluorescence background, and improve spectral fidelity [122]. These properties are useful for biofluid analysis, where weak molecular fingerprints are often masked by background interference. Nevertheless, carbon-based interfaces may also introduce nonspecific adsorption and surface heterogeneity, especially in protein-rich biological samples. In some cases, enhanced SERS signals may mainly originate from improved molecular enrichment rather than intrinsic chemical enhancement. Therefore, when evaluating Au-carbon SERS platforms, it is important to distinguish molecular adsorption effects from interfacial charge-transfer contributions and to assess whether the platform maintains selectivity and reproducibility in real biological matrices.

Overall, for EM-CM synergistically enhanced SERS substrates constructed from gold and other materials, the key factor is whether the second component can exert a substantive influence on the detection interface of the Au substrate. In the studies discussed in this section, these composites mainly complement the electromagnetic enhancement of Au through the regulation of interfacial electronic structure, surface modifiability, molecular adsorption, and functional response behavior. Despite differences in material systems, structural designs, and application targets, these studies consistently demonstrate that when the second component can participate in interfacial interactions or the detection process, the performance and applicability of gold-based SERS platforms can be further improved. Therefore, designing gold-based composites around EM-CM synergistic enhancement is an important strategy for constructing high-performance composite SERS substrates.

3.3. CM-Dominated Au-Based SERS Systems

Unlike conventional AuNPs, gold clusters and gold single atoms have sizes further reduced to the atomic or sub-nanometer scale, and their electronic structures and optical responses therefore undergo significant changes [123,124]. For traditional gold nanostructures, pronounced SERS enhancement usually relies on the strong localized electromagnetic fields generated by localized surface plasmon resonance, namely the typical electromagnetic enhancement mechanism [31]. However, when the size of gold is reduced to the cluster or even single-atom level, the system generally no longer possesses the continuous electronic characteristics and spatial scale required to form classical plasmon resonance and hotspot electric fields, and therefore it is difficult to provide electromagnetic enhancement comparable to that of conventional AuNPs [125–127]. Consequently, the enhancement in such systems mainly arises from interfacial charge transfer after molecular adsorption, reconstruction of local electronic states, and the resulting changes in dipole moment and polarizability, making them more suitable to be discussed as emerging gold-based Raman enhancement platforms dominated by CM [128,129].

From a theoretical standpoint, the enhancement behavior of gold clusters no longer simply follows the hotspot logic of large-sized metallic nanostructures, but is more strongly governed by electronic transitions at the molecule-cluster interface. You et al. systematically investigated the chemical enhancement behavior of SERS in complexes formed between para-substituted thiophenol derivatives and the gold cluster Au₁₃ using density functional theory (DFT) calculations (Figure 3A) [130]. In this work, Au₁₃ was employed as a microscopic model of the gold surface to compare the effects of different substituents and adsorption configurations on Raman enhancement. The results showed that, for small-sized gold clusters, the core of the enhancement is more closely related to the regulation of interfacial electronic structure rather than the electric field amplification brought about by nanohotspots.

This understanding has been further supported by more recent theoretical studies. Boto et al. proposed a theoretical computational framework for separating the contributions of chemical enhancement and electromagnetic enhancement in SERS, and applied it to a model system in which a biphenyl dithiolate (BPDT) molecule was positioned between two groups of gold clusters (Figure 3B) [131]. In this work, DFT was used to calculate the Raman polarizability of the molecule-gold cluster complex, time-dependent density functional theory (TDDFT) was employed to calculate the localized electromagnetic field induced by the gold clusters, and a Raman

scattering treatment under nonuniform fields was further introduced, thereby allowing the total EF, chemical EF, and electromagnetic EF to be obtained separately. The study showed that in small-sized gold cluster systems containing only 1, 4, 10, and 20 gold atoms on each side, the total SERS enhancement could reach the order of about 10^3 , and was mainly dominated by chemical enhancement, while electromagnetic enhancement contributed only as a minor auxiliary factor.

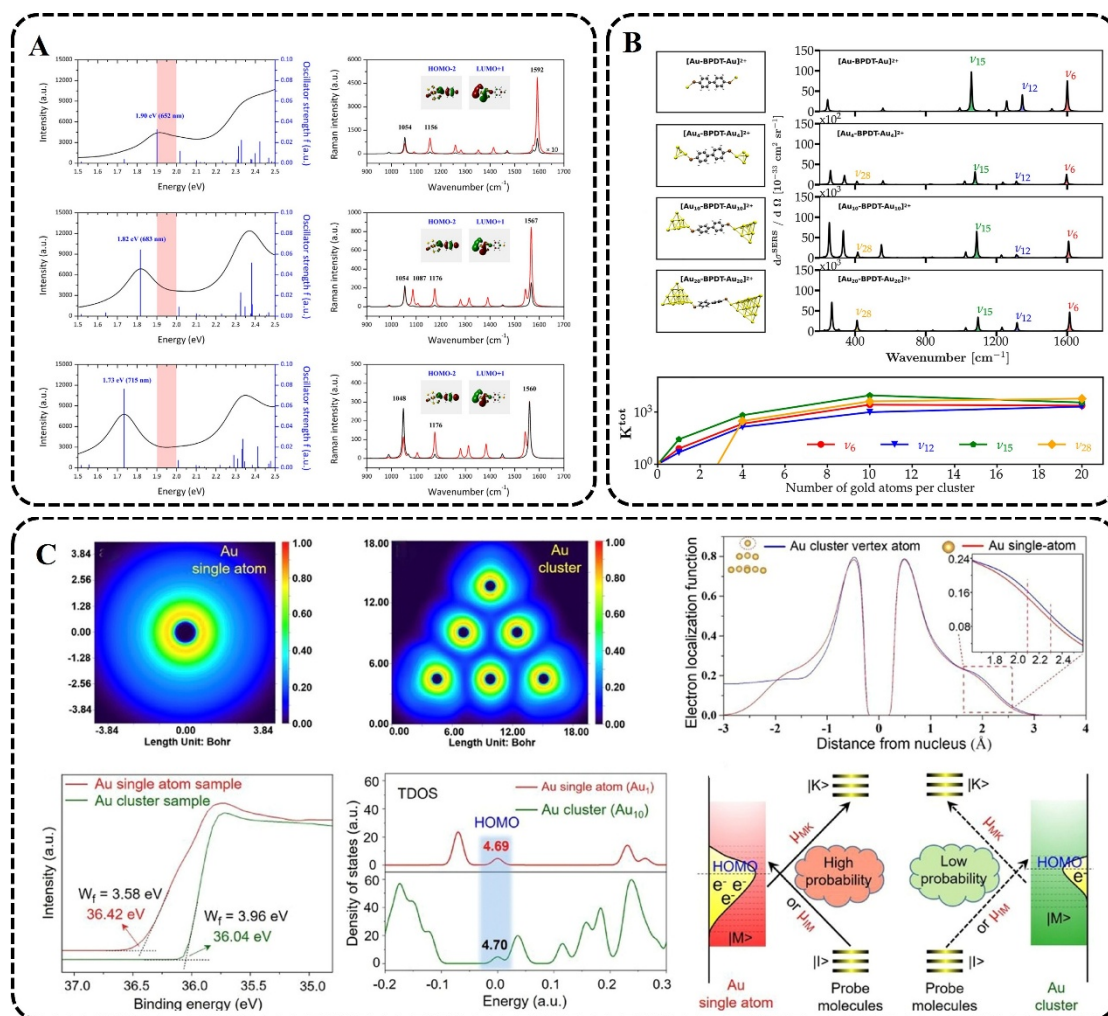


Figure 3. (A) Calculated excitation spectra and frequency-dependent Raman spectra for bridge-type OH-BT/Au13, SH-BT/Au13, and SeH-BT/Au13 complexes, illustrating the orbital characteristics involved in the primary charge-transfer transitions. Adapted with permission from Ref. [130]. Copyright 2016, Elsevier. (B) Minimum-energy structures, Stokes differential Raman cross-sections, and the evolution of the total SERS EF with gold cluster size for $[\text{Au}_n\text{-BPDT-Au}_n]^{2+}$ ($n = 1, 4, 10, 20$) complexes. Adapted with permission from Ref. [131]. Copyright 2024, American Chemical Society. (C) Electron localization function, ultraviolet photoelectron spectroscopy, total density of states, and energy-level scheme analyses for Au single atoms and Au cluster vertex atoms. Adapted with permission from Ref. [132]. Copyright 2022, American Chemical Society.

While gold clusters still retain certain characteristics of atomic assemblies, gold single atoms represent an even more extreme size scale. Yu et al. constructed a gold single-atom SERS substrate anchored on amorphous C_3N_4 nanosheets (Au_1/ACNs) and proposed the concept of single-atom-enhanced Raman scattering (SAERS) (Figure 3C) [132]. This system utilized the abundant defects and unsaturated coordination sites in amorphous C_3N_4 to stabilize Au single atoms, thereby preventing Au from aggregating into clusters. Unlike traditional gold-based SERS substrates that rely on plasmonic hotspots, this platform mainly enhanced molecular dipole moments and polarizability through interfacial charge transfer synergistically induced by Au single atoms and C_3N_4 . The study showed that Au_1/ACN could achieve an EF of 2.5×10^4 with only about 2.5% Au coverage area within the laser irradiation region, while also exhibiting excellent spectral stability and reproducibility. This work indicates that when gold further exists in the form of single atoms, the system no longer possesses the structural basis required to form conventional metallic nanohotspots, and its enhancement mechanism therefore more clearly points to

interfacial charge transfer and local electronic state regulation. Therefore, gold single atoms can be regarded as a more typical type of chemically enhanced gold-based substrate.

Overall, gold clusters and gold single atoms can be considered as an independent category in this chapter not only because of their smaller structural scale, but because this size reduction brings about a clear difference in the enhancement mechanism. Unlike conventional AuNPs, which mainly rely on localized surface plasmon resonance and hotspot electric fields, gold clusters and gold single atoms generally depend more on interfacial charge transfer after molecular adsorption, electronic state reconstruction, and the resulting changes in polarizability to achieve Raman enhancement. Existing theoretical and experimental studies have both shown that such systems are better understood as emerging gold-based Raman enhancement platforms characterized mainly by chemical enhancement, and therefore they are of considerable research value for expanding the structural boundaries of gold-based substrates and deepening the understanding of chemical enhancement mechanisms.

Although Au clusters and Au single atoms provide important model systems for understanding chemically dominated Raman enhancement, their biomedical applicability remains at an early stage. Compared with plasmonic Au nanoparticles, these ultrasmall Au species usually lack strong electromagnetic hotspots, and their overall enhancement capability may be lower for many practical sensing tasks. Their main value currently lies in clarifying interfacial charge-transfer mechanisms, active-site effects, and molecular adsorption-dependent enhancement. For translation into biomedical detection, future work needs to address signal intensity, substrate stability, biocompatibility of the supporting matrix, molecular selectivity, and validation in complex biological samples. Therefore, these systems should be regarded more as emerging mechanistic platforms than as mature diagnostic tools at the current stage.

Finally, to provide a clear quantitative overview of the representative gold-based SERS platforms discussed above, we summarized the EF, LOD, reproducibility, and biomedical applications for each system on Table 1.

Table 1. Quantitative comparison of representative Au-based SERS platforms.

Platform Category	Representative Examples	EF	LOD	Reproducibility	Biomedical Application	Ref.
Colloidal Au-based	Gold nanostar	/	/	/	In vivo early liver fibrosis detection in mice	[59]
	DNA origami Au nanorod dimer	/	0.2 nM	/	Label-free EGFR detection	[62]
	CRISPR/Cas12a-mediated AuNP aggregation SERS platform	/	pM level	/	HPV16/HPV18 nucleic acid detection	[63]
Solid Au-based	Au micro/nano ordered array	3.26×10^7	17 ppb	RSD = 2.83%	Etomidate detection	[74]
	Au nanocone array		/	RSD < 3.74%	Gastric/colorectal cancer biomarker detection	[75]
Core-shell Au-based	Ag@Au porous nanocage	/	0.12 ng mL ⁻¹	/	AFP sandwich immunoassay	[84]
	Au-Ag-Ag core-shell-shell nanosea cucumbers	/	26 particles μL^{-1} (LNCaP); 72 particles μL^{-1} (SKBR); 35 particles μL^{-1} (HepG2)	/	Multiplex exosome detection	[86]
Au-based fiber optic SERS system	Au-nanostructured fiber SERS optrode / lab-on-fiber platform	/	1×10^{-11} M (model analyte)	RSD=8.5%	/	[91]
Au-semiconductor composite	Rose-like MoS ₂ /Au nanocomposite	8.52×10^6	2.1×10^{-9} M (crystal violet)	/	/	[112]
	Au-ZnO hybrid nanoparticles	/	/	/	/	[110]
Au-carbon composite	GO-gold nanostar paper	/	0.436 μM (bilirubin)	/	Detection of free bilirubin in serum	[120]
Single atom-based	Au single atoms anchored on amorphous C ₃ N ₄	2.5×10^4	/	/	/	[132]

Note: “/” indicates that the corresponding value was not explicitly reported in the cited study. EF, enhancement factor; LOD, limit of detection; RSD, relative standard deviation; AFP, alpha-fetoprotein; EGFR, epidermal growth factor receptor; HPV, human papillomavirus; GO, graphene oxide; PET, polyethylene terephthalate; PS, polystyrene.

4. Biomedical Applications of Au-Based SERS Platform in Major Diseases Diagnosis

As discussed above, Au-based SERS substrates have evolved from signal-enhancing materials into functional analytical platforms through morphology engineering, assembly construction, interfacial modification, and EM-CM synergistic design. These advances enable the integration of target recognition, signal amplification, and scenario adaptability, thereby supporting their use in complex biomedical detection [47,133]. On this basis, Au-based SERS probes have gradually developed from simple signal-enhancing materials into functional analytical platforms that integrate target recognition, signal amplification, and scenario adaptability [134]. The practical value of these materials ultimately needs to be demonstrated through specific applications.

From a task-oriented perspective, different Au-based SERS platforms are suited to different biomedical analytical scenarios. Colloidal and surface-functionalized Au probes are particularly useful for liquid-biopsy-related targets, such as exosomes, miRNAs, and circulating tumor cells, because their good dispersibility and flexible surface chemistry facilitate interactions with analytes in solution and allow the introduction of antibodies, aptamers, nucleic acid probes, or antifouling layers [135–137]. Ordered solid Au substrates are more suitable for serum fingerprinting and complex biofluid discrimination, where stable interfaces, spatially uniform hotspots, and reproducible spectral output are required. Core-shell SERS tags and multiplex-encoded nanoparticles are more appropriate for immunoassays, tissue imaging, and intraoperative navigation because they can protect Raman reporters, provide high signal brightness, and support multiplexed readout [138,139]. In addition, EM-CM synergistic systems may offer extra advantages in complex biological environments through interfacial adsorption regulation, charge-transfer enhancement, background suppression, and multifunctional sensing. In the specific disease diagnosis cases discussed in this section, the relationship between material design features and analytical tasks can be more directly demonstrated [140].

At present, Au-based SERS probes have been widely used for the detection and analysis of biomarkers associated with various major diseases, among which high-burden diseases such as cancer, cardiovascular and cerebrovascular diseases, and neurological disorders are the most representative [26]. These diseases are of particular interest because they not only have high mortality or disability rates, but also generally share common features such as inconspicuous symptoms at the early stage, complex disease progression, and an urgent need for highly sensitive detection technologies. Among them, cardiovascular disease remains the leading cause of death worldwide, and cancer is also one of the major causes of death globally. Meanwhile, neurological disorders occupy an extremely important position in the global burden of disease and disability [13,14,141].

This section will focus on biomedical diagnostic applications and systematically summarize the research progress of Au-based SERS probes in the above three types of diseases.

4.1. Cancer Diagnosis

Cancer detection is one of the most active and representative application fields of gold-based SERS probes [142]. Research has gradually moved beyond the highly sensitive detection of single tumor biomarkers, but has gradually expanded to cell phenotyping analysis and intraoperative diagnostic support at the tissue level [143]. In terms of detection targets, related studies can generally be divided into three categories: tumor cell secretions, tumor cells, and tumor tissues. These applications demonstrate the potential of gold-based SERS probes in early cancer screening, tumor subtyping, and real-time diagnosis [28,135].

4.1.1. Detection of Tumor Secretions

In the detection of tumor secretions, exosomes and miRNAs are currently the two most intensively studied targets for gold-based SERS probes [144]. The former can reflect the secretory state and molecular composition of tumor cells, whereas the latter often serves as a nucleic acid biomarker associated with tumor occurrence and progression, participating in diagnosis and therapeutic evaluation [145,146]. The research focus in this area lies, on the one hand, in improving the sensitivity and specificity for detecting low-abundance biomarkers, and on the other hand, in achieving more stable and discriminative signal readout in complex biological samples [135,147].

Some studies have focused on the label-free analysis of the overall fingerprint information of exosomes. Guo et al. developed a label-free SERS exosome detection platform combined with thermophoretic enrichment and applied it to the in situ analysis and classification of cancer-related exosomes (Figure 4A) [148]. This system used a 785 nm laser to generate a temperature gradient on a superhydrophobic surface, enabling exosomes and Au nanoparticles to be co-enriched within 10 min into a small area of about 500 μm . As a result, the probability of interaction between exosomes and the hotspot regions of AuNPs was significantly increased. This also helped overcome the limitation that their relatively large size restricts direct access to conventional SERS hotspots. The

study showed that the platform achieved detection limits of 10^2 – 10^3 particles/ μL for exosomes derived from normal cells, hepatocellular carcinoma cells, and human serum, with signals enhanced by about two orders of magnitude compared with the natural drying method. Furthermore, when combined with principal component analysis (PCA) and machine learning analysis, the platform enabled effective discrimination between normal and cancer-related exosomes, with both sensitivity and specificity reaching 100%. This type of study emphasizes improving the feasibility of label-free exosome-based cancer classification through enrichment strategies and pattern recognition, rather than simple molecular quantification.

Other researchers have focused on the detection of miRNA in exosomes. Kang et al. constructed a closely packed gold octahedral array SERS sensing platform and applied it to the ultrasensitive detection of let-7a miRNA in exosomes derived from breast cancer cells (Figure 4B) [149]. In this system, the gold octahedra were uniformly oriented with their triangular facets facing downward, forming larger-area and more uniformly distributed “hot surfaces,” thereby improving the sensitivity and reproducibility for detecting low-abundance cancer biomarkers. On this basis, the authors achieved accurate quantification of let-7a in MCF-7 exosomes by using the conformational transformation of probe DNA from a hairpin structure to a double-stranded form. This work demonstrates that, for low-abundance nucleic acid biomarkers in cancer liquid biopsy, the combination of ordered gold substrates with nucleic acid recognition strategies can simultaneously provide high sensitivity and high specificity.

Some researchers have also aimed to achieve the simultaneous detection of multiple miRNAs using SERS probes. Si et al. constructed a SERS sensing array based on catalytic hairpin assembly (CHA) and applied it to the simultaneous detection of multiple cancer-related miRNAs, including miR-21, miR-221, miR-133a, and miR-1246 [150]. In this system, an Au/Ag alloy nanoparticle layer was immobilized at the bottom of four independent detection units, and AuAgNP SERS tags simultaneously modified with hairpin DNA and the Raman reporter molecule 4-mercaptobenzonitrile (MPBN) were introduced. When target miRNAs were present, the corresponding CHA cycling reaction was triggered, causing a large number of SERS tags to be anchored onto the surface of the corresponding detection units and to form abundant hotspots with the underlying AuAgNP layer, thereby significantly enhancing the Raman signal. The study showed that this platform could achieve highly specific simultaneous detection of four cancer-related miRNAs in the same sample. Compared with single-target nucleic acid detection, this type of array-based design better highlights the practicality of combined biomarker analysis and is also more consistent with the need for multi-indicator synergistic discrimination in cancer molecular diagnosis.

Overall, Au-based SERS probes are not only suitable for the ultrasensitive analysis of single targets in the detection of tumor cell secretions, but are also applicable to the combined discrimination of overall exosomal fingerprints and multiple miRNAs [151]. Moreover, the strategies adopted by researchers differ according to the analytical target: for specific nucleic acids inside exosomes, greater emphasis is placed on combining ordered gold substrates with nucleic acid recognition to improve sensitivity; for overall exosome analysis, more emphasis is placed on enrichment and label-free fingerprint readout; and for multi-target miRNAs, greater emphasis is placed on highly specific recognition [135]. Taken together, these studies indicate that the advantages of gold-based SERS platforms in cancer liquid biopsy are reflected not only in sensitivity, but also in their ability to accommodate multiple types of analytical demands [152].

4.1.2. Tumor Cell Detection and Phenotyping

Compared with the detection of tumor cell secretions, the analysis of circulating tumor cells (CTCs) more directly reflects tumor metastasis and disease progression, and is therefore also one of the important application directions of gold-based SERS probes in cancer diagnosis [153–156]. The key issue in this type of research is not merely cell detection, but how to achieve efficient identification, enrichment, quantification, and subsequent analysis of rare cells in the complex background of blood [157]. Therefore, related studies are often centered on the substrate morphology optimization, cell capture and enrichment, and multifunctional signal readout [12,158–160].

Wu et al. constructed three SERS-active probes based on different gold nanomorphologies and applied them to the direct detection of circulating tumor cells in blood without pre-enrichment [161]. In this system, spherical AuNPs, gold nanorods, and gold nanostars were used as the cores, respectively. 4-MBA was employed to provide the Raman signal, while rBSA-FA was used to achieve antifouling performance and folate receptor-targeted recognition. The study showed that all three types of probes could achieve highly specific CTC detection in complex blood samples, among which AuNS-MBA-rBSA-FA performed the best, with a detection limit as low as 1 cell/mL, which was significantly better than the 3 cells/mL achieved by the spherical and rod-shaped gold probes. This work indicates that, in CTC detection, the morphological optimization of gold nanostructures not only affects the hotspot effect, but also directly influences the detection sensitivity and reliability in complex samples.

How to achieve analyte enrichment while maintaining strong signal enhancement capability is also one of the key research focuses. Xue et al. constructed an improved SERS-active magnetic nanoparticle system (SPION-PEI@AuNPs-MBA-rBSA-FA) and applied it to the highly sensitive medical detection and magnetic separation of circulating tumor cells [159]. In this system, superparamagnetic iron oxide nanoparticles were used as the magnetic core, and AuNPs were assembled in situ on their surface to form a SERS-active interface. MBA was further modified as the Raman reporter molecule, and folic acid-conjugated rBSA was grafted to recognize folate receptor-positive tumor cells. Therefore, this gold-based platform could simultaneously achieve targeted capture of tumor cells, magnetic enrichment, and SERS quantitative readout. The study showed that this method could detect HeLa cells in blood with a detection limit as low as 1 cell/mL, and the SERS intensity exhibited a good linear relationship with cell concentration.

In addition to improving the capability of SERS substrates themselves, combining SERS with fluorescence and other detection methods for dual-modal or multimodal detection has also broadened the research direction. Some researchers constructed a SERS–fluorescence dual-modal CTC detection platform based on multifunctional gold nanomaterials and applied it to the specific capture, release, and highly sensitive detection of breast cancer-related circulating tumor cells (Figure 4C) [162]. In this system, an aptamer-modified AuNFs/ITO substrate was used as the cell-capture interface, taking advantage of its large specific surface area and PEG antifouling layer to improve the selective enrichment of CTCs. Meanwhile, the authors further designed AuNS signal probes modified with anti-EpCAM and HCR DNA, enabling dual-modal analysis of CTCs through a dual-recognition mechanism and multiple signal amplification. The study showed that, in the SERS mode, the platform exhibited a detection range of 5–200 cells/mL for MCF-7 cells, with a detection limit of 5 cells/mL; in the fluorescence mode, the detection range was 10–200 cells/mL, with a detection limit of 10 cells/mL, and the captured cells could be efficiently released for subsequent analysis.

Overall, in CTC detection, gold-based SERS probes can improve the sensitivity for rare-cell detection by optimizing gold nanomorphology and hotspot structures; they can enhance the enrichment efficiency of cells in complex blood samples by combining magnetic separation or capture interfaces; and they can also be further integrated with other detection signals such as fluorescence to expand the analytical potential of CTC detection [143].

4.1.3. Tumor Tissue Imaging and Intraoperative Diagnosis

In applications at the cancer tissue level, the value of gold-based SERS probes is mainly reflected in rapid imaging of fresh tissue surfaces, margin assessment, and intraoperative molecular guidance [163]. Compared with biofluid or cellular detection, this type of research deals with more complex and larger-scale samples, and therefore places greater emphasis on spatial localization capability and compatibility with clinical workflows [164,165].

Wang et al. proposed a quantitative molecular phenotyping method based on targeted gold-core SERS nanoparticles and applied it to the rapid margin assessment of freshly resected tissue surfaces during breast-conserving surgery (Figure 4D) [163]. In this system, gold-core SERS nanoparticles with different Raman barcodes were used as multiplex molecular probes. By targeting tumor-related receptors such as EGFR or HER2, and simultaneously introducing isotype control nanoparticles, the authors established a targeted/nontargeted ratio imaging strategy, thereby reducing interference caused by nonspecific adsorption and uneven delivery. This platform could complete staining and imaging within 15 min and perform rapid detection of fresh tissue surfaces with an area of about 4 cm² at submillimeter resolution. Its molecular imaging results were highly consistent with those of flow cytometry and immunohistochemistry, and it could be used to distinguish HER2-positive tumors from normal breast tissue. This work indicates that gold-based SERS nanoparticles can be applied not only in liquid biopsy, but also in intraoperative medical detection.

Similarly, Czaja et al. proposed a Raman surface topography imaging method based on targeted gold-core SERS nanoparticles and applied it to whole-surface intraoperative margin assessment of freshly resected specimens during breast-conserving surgery [165]. This system also used gold-core SERS nanoparticles as multiplex molecular probes, and by changing the reporter molecules and antibodies, it could simultaneously identify multiple tumor-related biomarkers. Furthermore, by combining a programmable rotating accessory with the microscope autofocus function, the platform enabled three-dimensional topographic Raman scanning of the entire resected specimen surface, thereby spatially locating potential residual tumor regions. The study showed that the platform could correctly identify and deconvolute 26 different SERS nanoparticles, achieved a linear correlation coefficient of 0.97 for quantitatively diluted signals, and completed the entire intraoperative imaging process within about 1 h. This work further demonstrates the spatial navigation potential of gold-based SERS nanoparticles in tumor surgery. They can be used not only for molecular-level identification, but also in combination with three-dimensional surface imaging to support more comprehensive intraoperative margin

assessment. Although tissue-level SERS imaging represents a more clinically relevant direction than many buffer-based sensing assays, its translational status should still be assessed cautiously. Targeted SERS nanoparticles can provide multiplexed molecular information and spatial maps on freshly resected tissues, making them promising for intraoperative margin assessment. However, exogenous probe staining, washing, spectral scanning time, nonspecific adsorption, and tissue heterogeneity may complicate routine surgical workflow. Larger prospective studies are still needed to determine whether this strategy can provide diagnostic benefits beyond standard histopathology or immunohistochemistry. Therefore, this approach is best viewed as a translationally promising technology rather than a fully mature clinical tool.

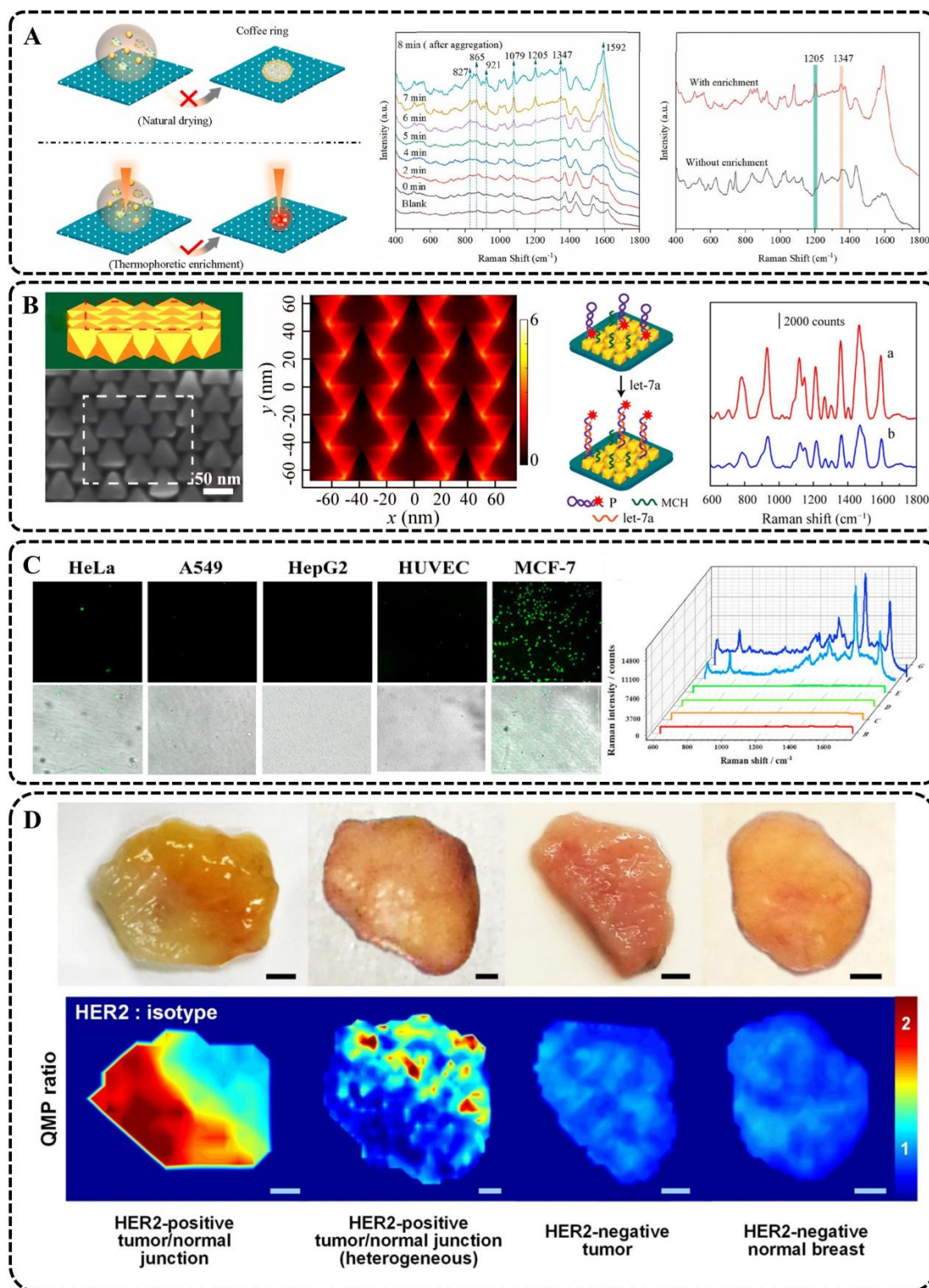


Figure 4. (A) SERS spectra of 293 exosomes (concentration: 106 particles/ μL) at varied laser irradiation times, a comparison between the natural drying and thermophoretic enrichment methods, and a schematic of the enrichment and non-enrichment procedures. Adapted with permission from Ref. [148]. Copyright 2024, Elsevier. (B)

Simulation model and SEM image of the Au octahedra array with electric field distribution profiles of 633 nm at the top Au/water interface, and schematic of the SERS sensing principle for let-7a with corresponding spectra in the absence and presence of 1 pM let-7a. Adapted with permission from Ref. [149]. Copyright 2021, American Chemical Society. (C) Fluorescence images demonstrating the capture specificity for different cell types (HeLa, A549, HepG2, HUVEC, MCF-7) on an aptamer-modified AuNFs substrate, and the corresponding Raman signals of the captured cells spiked with EpCAM-negative cancer cells (HeLa, A549, HepG2) and normal endothelial cells (HUVECs). Adapted with permission from Ref. [162]. Copyright 2021, Elsevier. (D) Photographs of HER2-positive and HER2-negative tissue specimens from four patients, and the corresponding concentration ratio images of HER2-NPs versus isotype-NPs. Adapted with permission from Ref. [163]. Copyright 2016, Springer Nature.

The role of gold-based SERS probes in cancer detection has further expanded from highly sensitive molecular sensing platform to that of a clinical auxiliary tool capable of supporting rapid imaging, margin assessment, and spatial navigation. This development does not represent a simple progression from the previously discussed liquid biopsy and cellular detection, but rather reflect the unique value of gold-based SERS technology in cancer diagnosis: it can not only identify tumor-related molecules and cells at the microscopic level, but also provide support for surgical decision-making at the macroscopic tissue scale.

4.2. Neurological Diseases Diagnosis

Neurological disorders are highly heterogeneous and include Alzheimer's disease (AD), Parkinson's disease (PD), and other neurodegenerative diseases [166]. Among them, AD has attracted substantial research attention and has developed a relatively mature molecular biomarker system. In particular, amyloid- β (A β), tau, and their related ratio indicators have relatively clear diagnostic significance in biofluid testing [167–171]. On this basis, the following section will take Alzheimer's disease as the main representative to summarize and discuss the applications of gold-based SERS substrates in the detection of neurological disorders.

4.2.1. Quantitative Detection of Neurological Biomarkers

For neurodegenerative diseases, the sensitive detection of low-abundance biofluid biomarkers remains a major focus in early diagnosis and risk assessment [172]. Improving the detection sensitivity and reproducibility of classical neurological disease biomarkers, as well as achieving multiplex analysis of multiple indicators in the same sample, are key issues that need to be addressed in this field [173].

For the highly sensitive quantitative detection of classical AD biomarkers, Shim et al. constructed a multiplex digital detection platform based on concave-convex core-shell gold nanoscale SERS probes and applied it to the simultaneous quantitative analysis of A β 42 and A β 40 (Figure 5A) [174]. This system possessed both "internal hotspots" and "external hotspots," significantly improving the intensity and reproducibility of single-particle SERS signals. Furthermore, the authors selected trifluoromethyl benzenethiol (TFMBA) and 4-MBA signal molecules with different Raman fingerprints to correspond to A β 42 and A β 40, respectively, thereby enabling crosstalk-free simultaneous detection of these two neurological disease biomarkers. The study showed that the detection limits of this platform for A β 42 and A β 40 were as low as 8.7×10^{-17} g/mL and 1.0×10^{-15} g/mL, respectively, and the detection range covered clinically relevant concentration ranges in cerebrospinal fluid and plasma. This work demonstrates that gold-based SERS nanoprobe with finely engineered structures can greatly improve the sensitivity of neurological disease biomarker detection, thereby showing strong potential for early biofluid diagnosis of AD.

Compared with dual-marker detection, multiplex biomarker analysis is more consistent with the practical needs of clinical risk assessment for neurodegenerative diseases. Muhammad et al. constructed a porous glass micropillar chip platform based on aptamer-SERS nanoprobe and applied it to the combined blood-based detection of multiple biomarkers for the early diagnosis of Alzheimer's disease (Figure 5B) [175]. In this system, magnetic Fe₃O₄/Au capture probes and AuNR/Ag reporter probes were designed separately, and different Raman reporter molecules were used to achieve the simultaneous analysis of five AD-related proteins, namely neurogranin (Nrgn), angiopoietin-2 (Angio-2), peroxiredoxin 3 (PRDX3), L-lactate dehydrogenase (L-LDH), and tau-441 (τ -441). The study showed that the platform achieved detection limits in blood down to the attogram (ag) level, with LOD values of 24.8, 0.63, 0.34, 0.76, and 4.45 ag/mL for the five biomarkers, respectively. Furthermore, this method could be used for dynamic evaluation of disease progression in an AD mouse model and could distinguish neurological disease-related risk profiles among different age groups in human blood samples.

4.2.2. Recognition of Pathology-Related Aggregation States

In neurodegenerative diseases, the aggregation state of abnormal proteins is closely related to disease occurrence and progression. Therefore, quantifying total biomarker levels alone is often insufficient to fully reflect pathological progression. How to identify different states such as monomers, oligomers, and fibrils has gradually become another important application direction of gold-based SERS in neurological disease detection [176,177]. The focus of this type of research is not only to detect the presence of a certain molecule, but also to further characterize its pathology-related conformational changes and aggregation evolution process.

Wang et al. constructed a graphene oxide/gold nanoparticle (GO/Au NPs) nanohybrid SERS substrate and applied it to the ultrasensitive label-free detection of A β 1-42 monomers and fibrils, which are key biomarkers of AD [178]. In this system, Au nanoparticles were densely loaded onto the GO surface through in situ reduction, thereby simultaneously taking advantage of the plasmonic enhancement provided by Au nanoparticles, the molecular enrichment effect arising from the large specific surface area of GO, and the chemical enhancement effect, thus improving the detection capability for neurological disease-related protein biomarkers such as A β 1-42. The study showed that the platform achieved detection limits as low as 0.0232 ng mL⁻¹ and 0.0192 ng mL⁻¹ for A β 1-42 monomers and fibrils, respectively, and could further monitor the aggregation process of A β 1-42 from monomers to fibrils. This demonstrates promising application potential in the early diagnosis of neurodegenerative diseases.

Other researchers constructed a SERS detection platform based on floating gold microbubbles (Au/MB) and applied it to the sensitive detection and aggregation-state monitoring of AD-related A β 1-40 oligomers (Figure 5D) [179]. In this system, Au nanoclusters were grown in situ on the surface of polyvinyl alcohol air microbubbles, thereby constructing a gold-based SERS substrate with both near-infrared plasmonic enhancement and a three-dimensional curved detection interface. The authors employed a Cu²⁺/4-MBA functional design to convert the interaction between A β 1-40 and Cu²⁺ into changes in the conformation and orientation of 4-MBA molecules, thereby enabling detection through SERS frequency shifts and peak intensity changes. The study showed that this platform achieved a detection limit of 10⁻⁹ M for A β 1-40 oligomers preincubated for 48 h and could distinguish A β samples at different aggregation stages. At the same time, it exhibited good anti-interference performance in phosphate-buffered saline (PBS), Tris-HCl, and artificial cerebrospinal fluid, demonstrating its application potential in Alzheimer's disease detection. This aggregation-state sensing strategy is valuable because it moves beyond simple concentration measurement and attempts to capture pathological conformational changes of A β species. Such information is more closely related to neurodegenerative mechanisms than total biomarker concentration alone. However, its current biomedical applicability should still be viewed cautiously. The aggregation state of A β is highly sensitive to incubation time, buffer composition, metal ion concentration, and sample handling conditions. Therefore, signals obtained from preincubated A β oligomers in controlled solutions may not fully represent the complexity of A β species in patient cerebrospinal fluid or plasma. In addition, whether the observed spectral changes can be quantitatively correlated with disease stage or cognitive decline remains to be established. Thus, this platform is mechanistically meaningful and useful for studying A β aggregation, but further validation in clinically characterized samples is needed before it can be considered a diagnostic technology for AD.

The common significance of these studies lies in showing that gold-based SERS platforms in neurological disease analysis can not only serve as tools for conventional quantitative detection, but can also be used to identify key state differences during pathological aggregation processes. Compared with simple concentration detection, aggregation-state recognition is closer to the actual pathological process of diseases such as AD and better reflects the unique advantage of SERS in resolving molecular structural information.

4.2.3. Identification of Emerging Molecular Features

In addition to classical protein biomarkers and pathological aggregation states, the molecular abnormalities of neurological diseases may also be manifested as more fine-grained chemical feature changes, such as differences in molecular chirality or other more subtle structural characteristics [180,181]. Compared with traditional detection methods, the main advantage of gold-based SERS platforms in this direction lies in their ability to sensitively read out complex molecular features while retaining, to a certain extent, fingerprint information related to molecular structure, thereby supporting the discovery of novel biomarkers [134].

Hao et al. constructed a chiral gold nanostructure monolayer SERS substrate and applied it to the ultrasensitive detection and enantiomeric discrimination of chiral metabolic biomarkers associated with AD (Figure 5C) [182]. In this system, chiral gold nanostructures induced by D-/L-cysteine-leucine dipeptides were used as the basic units and were further assembled into a two-dimensional monolayer film, thereby simultaneously achieving interparticle electromagnetic coupling enhancement and chiral surface-selective adsorption capability.

Therefore, this platform could not only enhance the Raman signals of kynurenine (Kyn) and tryptophan (Trp), but also distinguish their D/L enantiomers. The study showed that the D-Au monolayer achieved detection limits of 3.6 nM and 4.1 nM for D-Kyn and D-Trp, respectively, while the L-Au monolayer achieved detection limits of 3.7 nM and 4.4 nM for L-Kyn and L-Trp, respectively. More importantly, the authors found in real samples that serum D-Kyn in AD patients was significantly higher than that in healthy controls, and that D-Kyn could be detected in cerebrospinal fluid samples only from AD patients. This work demonstrates that chiral gold-based SERS platforms can achieve enantiomeric discrimination capability and therefore have promising application value in the discovery of novel metabolic biomarkers for AD.

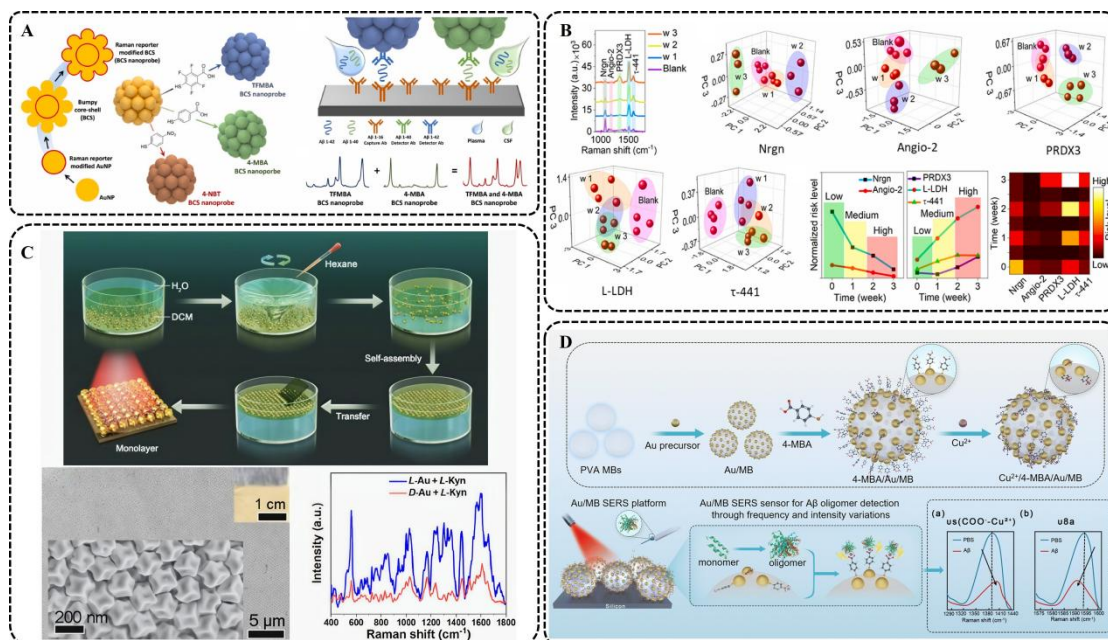


Figure 5. (A) A multiplexed digital sensing platform based on bumpy core-shell SERS nanoprobe synthesis, the sensing principle for the simultaneous detection of A β 40 and A β 42, and multiplexed analytical validation in a mixed sample. Adapted with permission from Ref. [174]. Copyright 2025, Elsevier. (B) Real-time SERS multiplexing of serum biomarkers with PCA-based quantification/classification and brain injury risk assessment via time-biomarker expression correlation. Adapted with permission from Ref. [175]. Copyright 2025, Elsevier. (C) Schematic of the chiral Au-CL monolayer fabrication process, SEM and optical micrographs of the L-Au-CL monolayer, and SERS spectra of L-kynurenine acquired on D- and L-Au-CL monolayers. Adapted with permission from Ref. [182]. Copyright 2025, John Wiley and Sone. (D) Schematic of the Cu²⁺ and 4-MBA functionalized buoyant plasmonic Au/MB SERS sensing substrate preparation and its application for oligomerized A β protein detection. Adapted with permission from Ref. [179]. Copyright 2025, Royal Society of Chemistry.

Although the number of such studies is still relatively limited at present, their significance lies not only in introducing a new category of detection targets. More importantly, they suggest that gold-based SERS research on neurological diseases is gradually expanding from the quantitative analysis of classical known biomarkers to the identification of more fine-grained molecular features [183]. Such strategies help overcome the limitations of relying solely on traditional protein biomarkers and also provide new ideas for subsequent studies on neurological diseases based on metabolic abnormalities and changes in molecular microstructure.

Current studies related to neurological diseases are no longer limited to the highly sensitive detection of single molecules, but have gradually expanded to dual-indicator and multi-biomarker combined analysis, recognition of pathological aggregation states, and the characterization of more fine-grained molecular features. This indicates that the value of gold-based SERS platforms in neurological disease detection lies not only in their sensitivity, but also in their ability to incorporate molecular fingerprint information, thereby providing richer analytical dimensions for early disease diagnosis and mechanism-related studies.

4.3. Cardiovascular Diseases Diagnosis

Cardiovascular diseases are among the leading causes of death worldwide, and their clinical diagnosis relies heavily on the rapid and accurate identification of key molecular biomarkers and pathological states [184,185]. Compared with other analytical techniques, the advantages of gold-based SERS substrates and probes in this field

are mainly reflected in rapid detection, high sensitivity, and the potential for integration into portable devices [186,187]. Therefore, current research applications in this area include the highly sensitive detection of single biomarkers, multiplex biomarker detection, and portable as well as point-of-care testing platforms.

4.3.1. Detection of Cardiovascular Disease Biomarkers

In the diagnosis of cardiovascular diseases, the highly sensitive quantitative detection of key biomarkers remains the most fundamental and also the most mature application direction [188]. In particular, biomarkers such as cTnI and B-type natriuretic peptide (BNP) are of great importance in clinical diagnosis and disease assessment. Gold-based SERS platforms are often used to detect such indicators, demonstrating their high sensitivity advantages in trace protein detection [189].

Xiang et al. constructed a sandwich-type SERS immunosensing platform based on an ordered multispiked gold nanostar array and applied it to the highly sensitive detection of cardiac troponin I (cTnI), a biomarker of acute myocardial infarction [190]. In this system, a uniform monolayer array of gold nanospheres was first prepared, followed by the formation of a multispiked gold nanostar array induced by 4-(2-hydroxyethyl)-1-piperazineethanesulfonic acid (HEPES), and capture antibodies were then immobilized on the surface of the gold nanostar substrate. In addition, Au nanolabels carrying 4-MBA and detection antibodies were constructed to form a sandwich immunostructure with cTnI, thereby enabling dual plasmonic enhancement detection. The study showed that this platform exhibited a good linear response toward cTnI in the range of 0.01–100 ng mL⁻¹, with a detection limit as low as 9.09 pg mL⁻¹, and showed good anti-interference performance against AFP, BSA, and horse serum, demonstrating its potential for the rapid in vitro diagnosis of acute myocardial infarction.

Similarly, Lin et al. constructed an aptamer-modified magnetic bimetallic SERS substrate (Fe₃O₄@Ag@Au-aptamer (Apt)) and applied it to the quantitative detection of cTnI, a biomarker of acute myocardial infarction (Figure 6A) [191]. In this system, Fe₃O₄ was used as the magnetic core, and Ag and Au were deposited layer by layer with the assistance of polydopamine (PDA), thereby forming a functionalized gold-based substrate that integrates magnetic separation capability, bimetallic Raman enhancement capability, and an Au-S aptamer immobilization interface. Subsequently, cTnI was specifically captured by the aptamer, and Coomassie Brilliant Blue G-250 (CBBG) was introduced as the Raman signal molecule to achieve quantitative readout of the target protein. This platform exhibited a good linear relationship for cTnI in the range of 0.01–100 ng mL⁻¹, with a detection limit as low as 5.50 pg mL⁻¹, while the recovery in spiked serum samples from healthy individuals ranged from 92% to 115%. This study more strongly highlights the integration of enrichment, recognition, and signal enhancement functions within a single platform, thereby improving the sensitivity and practical sample applicability of cardiovascular disease biomarker detection.

Overall, this type of research mainly centers on the precise quantification of key biomarkers, with the focus on improving detection sensitivity and reliability in complex samples through the structural optimization of gold-based substrates. Such platforms constitute the fundamental application form of gold-based SERS in cardiovascular in vitro diagnostics.

4.3.2. Portable and Point-of-Care Testing SERS Platforms

Given the convenience required for testing cardiovascular disease patients and the rapid onset of certain cardiovascular conditions, another important application direction of gold-based SERS substrates in the cardiovascular field is their integration with devices such as optical fibers, microfluidics, and portable chips to develop analytical platforms suitable for point-of-care testing (POCT) [192,193]. Such studies usually emphasize detection speed, device miniaturization, simplified operation, and the potential for clinical on-site use. The focus has shifted from feasibility to practicality, emphasizing whether detection can be completed more quickly and more portably.

Dong et al. constructed a pump-free microfluidic chip sensing platform based on SERS and applied it to the ultrasensitive and rapid detection of creatine kinase-MB (CK-MB), a biomarker of acute myocardial infarction (Figure 6B) [194]. In this system, gold-shell magnetic particles (AuMNPs) were used as capture probes, while AuNP SERS tags loaded with malachite green isothiocyanate (MGITC) and detection antibodies were used as signal probes. Through sandwich immunorecognition, an “AuMNP/CK-MB/SERS tag” complex was formed and enriched in situ within the chip detection chamber under a magnetic field. Meanwhile, the PEG-modified polydimethylsiloxane (PDMS) microchannel could drive sample flow automatically by capillary force without the need for an external pump, making it more suitable for POCT applications. The study showed that this platform exhibited a good linear response toward CK-MB in the range of 1 pg/mL to 10 µg/mL, with a detection limit as low as 1 pg/mL, and the entire detection process could be completed within 15 min. It could also be applied to

serum sample analysis, demonstrating its application capability for the rapid in vitro diagnosis of acute myocardial infarction.

Campu et al. proposed a portable microfluidic plasmonic chip based on gold bipyramids (AuBPs) and applied it to the rapid real-time detection of cTnI (Figure 6C) [195]. By immobilizing self-assembled AuBPs on a glass substrate and integrating them with PDMS microfluidic channels, the authors constructed a gold-based detection platform that combined LSPR, SERS, and thermoplasmonic responses. The study showed that, in the thermoplasmonic mode, this platform exhibited linear responses toward cTnI within two clinically relevant ranges, 10–100 pg mL^{-1} and 0.5–2 ng mL^{-1} , with detection limits of 4.2 pg mL^{-1} and 0.47 ng mL^{-1} , respectively. Furthermore, when validated using 16 real plasma samples, it demonstrated 75% sensitivity and 100% specificity, and the detection could be completed within 5 min. This indicates that the platform is suitable for rapid on-site diagnosis of cardiovascular diseases such as acute myocardial infarction.

This indicates that the application of gold-based SERS platforms in cardiovascular diseases is extending from traditional laboratory testing toward portable, microfluidic, and point-of-care detection. Unlike approaches that focus solely on achieving lower detection limits, this type of work places greater emphasis on compressing the detection workflow, integrating devices, and improving on-site operability, and therefore represents an important pathway for advancing gold-based SERS toward practical clinical application.

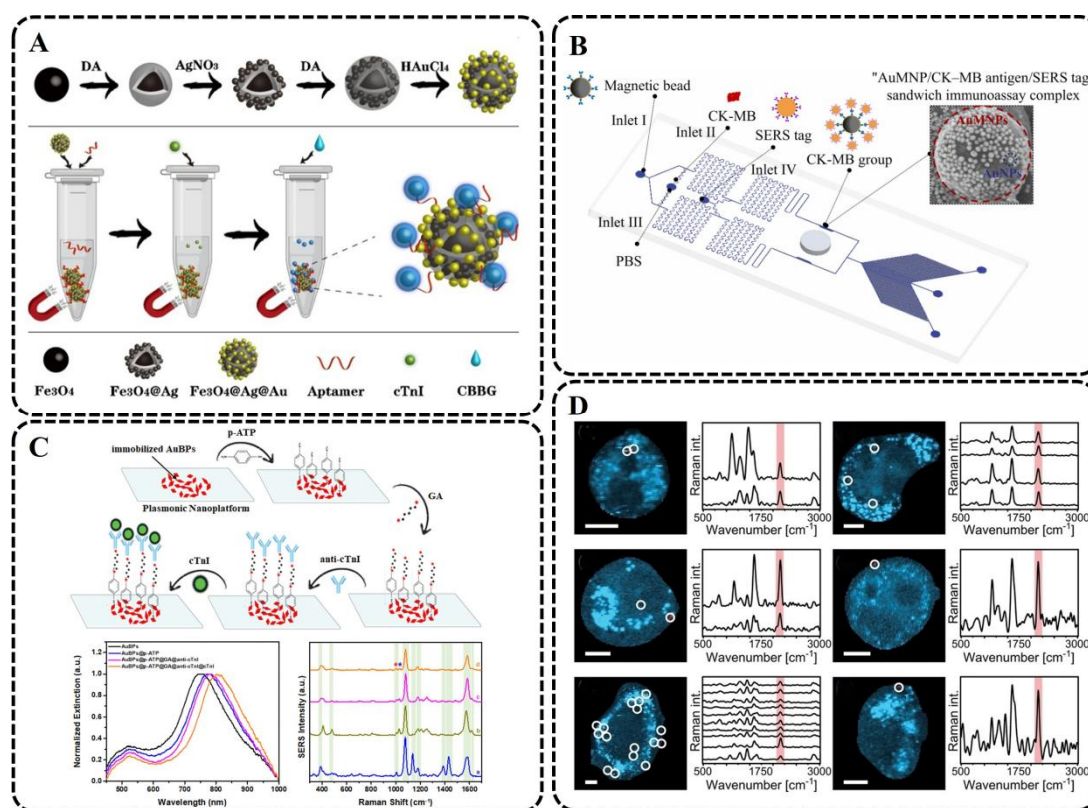


Figure 6. (A) Schematic diagram of the technical principle for the SERS capture probe (Fe₃O₄@Ag@Au-Apt). Adapted with permission from Ref. [191]. Copyright 2022, Springer Nature. (B) Schematic of the immunoassay procedure in a microfluidic chip. Adapted with permission from Ref. [194]. Copyright 2022, Elsevier. (C) Schematic of the biosensing protocol for cTnI detection, along with the extinction and corresponding SERS spectra acquired at each step, with characteristic vibrational bands marked. Adapted with permission from Ref. [195]. Copyright 2024, Royal Society of Chemistry. (D) Raman images of THP-1 macrophages incubated with AuNP@PDI@silica-man (six panels) and corresponding point spectra extracted from the red-marked locations in the images, showing the specific Raman reporter signal in all inspected cells. Adapted with permission from Ref. [196]. Copyright 2018, John Wiley and Sons.

4.3.3. Molecular Imaging and In Situ Visualization

In addition to biofluid biomarker detection, gold-based SERS probes have also been used for molecular imaging and in situ visualization analysis related to cardiovascular lesions. The goal is no longer limited to the quantification of a certain biomarker in blood, but rather to make use of the highly specific molecular readout

capability of SERS to spatially localize inflammatory cells, vascular adhesion molecules, and vulnerable plaque-associated microenvironments, thereby serving the risk assessment of diseases such as atherosclerosis.

Dugandžić et al. constructed a mannose-modified gold nanoparticle SERS probe and applied it to the targeted imaging of macrophages associated with vulnerable atherosclerotic plaques (Figure 6D) [196]. In this system, branched AuNPs were used as the SERS enhancement core, and a reporter molecule, perylene diimide (PDI), located in the Raman-silent region was further introduced. A silica coating was then applied to improve biocompatibility, and mannose was used to achieve specific recognition of the cluster of differentiation 206 (CD206) receptor on the surface of mature macrophages. The study showed that silica coating and mannose modification could significantly improve probe uptake by macrophages, while the SERS signal in the silent region helped achieve clear readout in complex cellular backgrounds. This work indicates that such probes can be used not only for the molecular recognition of inflammation-related cells, but also for targeted Raman imaging of vulnerable atherosclerotic plaques, thereby providing an imaging-based detection foundation for early risk assessment of cardiovascular diseases.

A set of antibody-functionalized gold-based SERS biofunctional nanoparticles have constructed and applied to in vivo multiplex molecular imaging of vascular inflammation [197]. In this system, AuNPs served as the SERS enhancement core, and different Raman reporter molecules together with targeting antibodies were used to recognize intercellular adhesion molecule-1 (ICAM-1), vascular cell adhesion molecule-1 (VCAM-1), and P-selectin, thereby enabling the simultaneous detection and discrimination of inflammation-related adhesion molecules associated with atherosclerosis. This platform could not only perform multiplex imaging and semiquantitative analysis on human endothelial cells, but also distinguish plaque regions from non-plaque regions in human coronary atherosclerotic tissues. Furthermore, the authors achieved noninvasive in vivo SERS molecular imaging after intravenous injection in a humanized mouse model and successfully detected vascular expression of ICAM-1 and P-selectin. This work indicates that gold-based SERS probes are not only suitable for the detection of cardiovascular disease biomarkers, but can also be extended to the molecular imaging of vascular inflammation and vulnerable plaques, showing promising clinical potential in the risk assessment of atherosclerosis. Compared with blood biomarker assays, SERS molecular imaging of vascular inflammation represents a more ambitious and clinically relevant direction because it aims to localize disease-associated molecular events within vascular lesions. The ability to detect ICAM-1, VCAM-1, and P-selectin in cells, human atherosclerotic tissues, and animal models suggests that targeted Au-based SERS nanoparticles can provide spatially resolved information that conventional soluble biomarker assays cannot offer. However, this approach also faces higher translational barriers. In vivo vascular imaging requires efficient probe delivery to lesion sites, sufficient penetration or optical access, low nonspecific retention, and rigorous safety evaluation of injected nanoparticles. Moreover, integration with clinically practical imaging instruments remains challenging, particularly for deep or moving vascular tissues. Therefore, this strategy has strong biomedical relevance as a molecular imaging concept, but its clinical translation will depend on probe safety, imaging accessibility, quantitative calibration, and compatibility with intravascular or endoscopic Raman systems

This type of research reflects another application value of gold-based SERS in cardiovascular diseases, namely its extension from “in vitro molecular detection” to “lesion-related molecular visualization.” Although the number of studies in this area is still clearly smaller than that on biofluid biomarker detection, the demonstrated spatial resolution capability and lesion-targeted recognition potential indicate that gold-based SERS is expected to play a more direct role in the risk assessment of vascular lesions such as atherosclerosis.

Overall, the applications of gold-based SERS in cardiovascular diseases can be divided into several scenarios. First, gold-based substrates can support the highly sensitive detection of key biomarkers, providing a foundation for the in vitro diagnosis of diseases such as acute myocardial infarction and heart failure. Second, the introduction of microfluidic, optical fiber, and paper-based devices has promoted the development of gold-based SERS toward portability and point-of-care testing. Third, molecular imaging and in situ visualization analysis have expanded the application boundaries of gold-based SERS in the identification of vascular inflammation and vulnerable plaques. These examples indicate that the value of gold-based SERS platforms in cardiovascular disease detection lies not only in their high sensitivity, but also in their ability to adapt to different diagnostic scenarios and practical needs.

5. Conclusions and Perspectives

5.1. Conclusions

Au-based SERS substrates have progressed from early enhancement-driven materials into increasingly sophisticated biomedical analytical platforms that integrate structural design, interfacial engineering, and

functional integration. From a material perspective, representative strategies such as morphology control, nanoaggregate construction and core-shell architectures, have defined a family of systems governed primarily by EM enhancement, where Raman signal amplification originates from localized surface plasmon resonance and hotspot-induced electric fields. More recently, the emergence of composite architectures and ultrasmall gold species has expanded the fundamental enhancement paradigm. By incorporating semiconductors, carbon materials, gold clusters, and gold single atoms, researchers have introduced interfacial charge-transfer interactions and adsorption-mediated regulation into SERS systems, leading to a transition from purely EM-dominated mechanisms to EM-CM synergistic regimes, and ultimately toward chemically dominated enhancement at the atomic scale.

At the application level, gold-based SERS platforms have demonstrated broad utility in biomedical detection, particularly in the analysis of biomarkers associated with cancer, cardiovascular diseases, and neurological disorders. These applications highlight their capability for ultrasensitive biomolecule detection, cellular phenotyping, tissue-level discrimination, and multiplexed biomarker analysis. Related studies have shown that gold-based SERS platforms exhibit significant potential in trace biomolecule detection, cell phenotyping, differentiation of diseased tissue regions, and multiplex biomarker analysis. Importantly, the field is undergoing a conceptual shift beyond the pursuit of maximal enhancement factors. Increasing emphasis is now placed on probe stability in complex biological environments, molecular recognition specificity, task-oriented adaptability, and compatibility with real biological systems. This reflects a transition from “highly sensitive substrates” to “task-oriented analytical platforms” tailored for specific biomedical applications.

Despite these advances, current progress remains largely confined to laboratory-scale material development and validation in simplified model systems. Critical challenges—including reproducibility, standardization, robustness in complex environments, and clinical translatability—remain unresolved. Therefore, addressing these limitations and defining clear development pathways will be essential for advancing Au-based SERS technologies toward practical biomedical implementation.

5.2. Challenges in Biomedical Applications

5.2.1. Reproducibility and Standardization

For medical detection, stable and reproducible signal output is essential for analytical results. However, SERS signals are highly dependent on the size, morphology, gap, and surface state of nanostructures, resulting in variations not only between batches but also across different regions of the same substrate. For colloidal systems, instability in particle aggregation state and dispersion behavior can directly affect hotspot distribution; for solid systems, minor fabrication variations may lead to signal nonuniformity. Therefore, how to establish more controllable synthesis and fabrication methods, improving the consistency of substrate preparation, and promote the standardization of testing developing controllable synthesis methods, improving preparation consistency, and standardizing testing and data analysis remain critical priorities.

5.2.2. Stability in Complex Biological Environments

Compared with ideal solution systems, real biological samples usually contain complex components such as proteins, salt ions, small-molecule metabolites, and other potential interfering components. These factors can affect the adsorption behavior of target molecules and may also lead to nonspecific signal enhancement, probe aggregation, or signal drift. This is particularly important for colloidal probes, which must maintain structural and interfacial stability in complex media such as blood, serum, urine, or tissue microenvironments. Although some studies have improved these issues through surface coating, internal standards, and interfacial modification, maintaining long-term performance and signal reliability in complex environments remains challenging.

5.2.3. Cost and Scalable Fabrication

Gold materials themselves are relatively expensive, and high-performance gold-based SERS substrates often rely on complex morphology control, precise assembly, multistep surface modification, and functional molecule conjugation. This not only increases fabrication cost, but also raises process complexity and the difficulty of large-scale production. For practical applications, the lack of scalable, low-cost production will hinder practical translation. At the same time, substrate reusability directly affects cost-effectiveness. Although some platforms have attempted to improve substrate reusability through strategies such as photocatalysis, self-cleaning, and reversible adsorption, the field remains at an early stage. In medical detection, this issue is related not only to

signal recovery performance, but also to risks such as contamination and cross-interference. Therefore, balancing cost, scalability, performance, and stability remains a key challenge for practical application.

5.2.4. Clinical Translation

At present, many studies still remain at the stage of simulated samples or small-scale sample validation, and there is still a considerable distance to true clinical translation. In addition, the lack of standardized protocols across instruments, sample preparation, and data analysis complicates result comparison and clinical evaluation. Therefore, advancing toward clinical use requires improvements in both materials and standardized workflows, data processing, and evaluation systems.

5.3. Future Perspectives

5.3.1. Cost-Effective Design with High Performance

Future development of gold-based SERS substrates should place greater emphasis on balancing material design with manufacturing cost. On one hand, the complexity of preparation can be reduced by developing simpler and higher-yield wet-chemical synthesis, self-assembly, and template-assisted fabrication methods. On the other hand, under limited cost conditions, it is still necessary to continue optimizing particle morphology, gap structures, and surface modification strategies in order to achieve the coordinated improvement of enhancement capability, stability, and biological applicability. For platforms truly oriented toward practical application, “high performance with manufacturability” will be more meaningful than simply pursuing extreme enhancement factors (Figure 7).

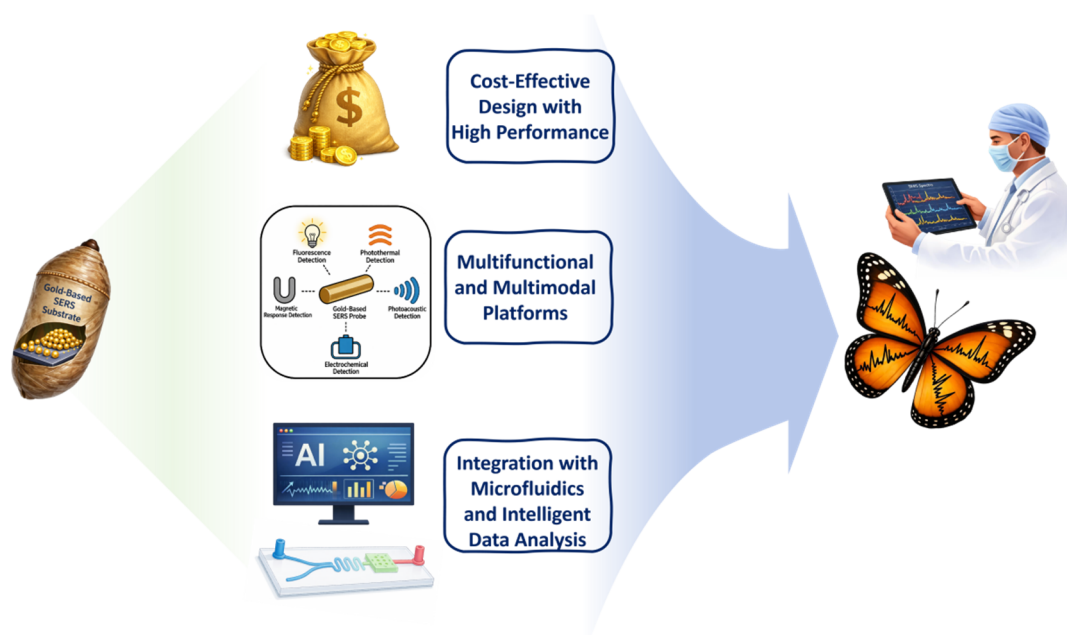


Figure 7. Prospects and future directions of gold-Based SERS substrates in biomedical detection.

5.3.2. Multifunctional and Multimodal Platforms

As analytical demands continue to increase, probe platforms with a single signal output are no longer sufficient to meet the requirements of complex biological system analysis. In the future, gold-based SERS probes are expected to be further integrated with functions such as fluorescence, photothermal effects, photoacoustic response, electrochemical properties, and magnetic responsiveness to form multimodal detection platforms. This can not only enable multidimensional information complementation and improve detection accuracy and reliability, but also help expand their application potential in *in vivo* imaging, dynamic monitoring, and therapeutic response evaluation. Particularly for complex disease systems, multimodal platforms show clear potential for improving diagnostic precision.

5.3.3. Integration with Microfluidics and Intelligent Data Analysis

The integration of microfluidic platforms with Au-based SERS systems has emerged as an important direction for biomedical sensing. Recent studies have demonstrated that microfluidic devices can provide controlled environments for sample processing, target enrichment, reaction regulation, and signal acquisition, while reducing sample consumption and assay time. In addition, microfluidic-assisted SERS platforms have shown advantages in improving analyte transport, enhancing target capture efficiency, and enabling continuous-flow, droplet-based, and multiplexed detection. Further integration of microfluidics with Au nanostructures, magnetic separation strategies, and immunoassays may facilitate the development of more automated and reproducible diagnostic workflows.

At the same time, advances in artificial intelligence and machine learning are reshaping the analysis of complex SERS datasets. These approaches have already been applied to spectral preprocessing, spectral classification, peak identification, feature extraction, and quantitative prediction, particularly in disease diagnosis, pathogen detection, cell phenotyping, and multiplex biomarker analysis. As SERS datasets continue to grow in complexity, intelligent algorithms are expected to play an increasingly important role in improving analytical accuracy and extracting clinically relevant information from spectral fingerprints.

Despite these advances, several challenges remain. Current studies are often limited by relatively small datasets, inconsistent preprocessing procedures, instrument- and substrate-dependent spectral variations, and insufficient external validation. Future efforts should therefore focus on standardized data acquisition protocols, high-quality spectral databases, interpretable learning frameworks, cross-platform calibration, and multicenter validation. The continued convergence of microfluidics, Au-based SERS technologies, and intelligent data analysis is expected to accelerate the translation of SERS from proof-of-concept studies toward robust, automated, and clinically deployable diagnostic platforms.

5.3.4. From “High-Enhancement Substrates” to “Task-Oriented Platforms”

Overall, the future development of gold-based SERS substrates will shift from pursuing higher enhancement factors to optimizing overall performance for specific tasks. This means that material design needs to simultaneously consider signal intensity, interfacial recognition, sample adaptability, detection workflow, and application scenarios. In other words, the true maturity of gold-based SERS technology lies not in pushing a single performance metric to the extreme, but in developing platforms that combine high sensitivity, high reliability, and high adaptability for real biomedical applications.

Author Contributions

S.Z.: Writing—original draft, Writing—review & editing, Investigation, Formal analysis. X.C.: Writing—review & editing, Investigation, Visualization. L.S.: Writing—review & editing, Resources, Methodology. X.Y.: Writing—review & editing, Supervision. A.W.: Writing—review & editing, Supervision, Funding acquisition. J.L.: Writing—review & editing, Supervision, Funding acquisition. All authors have read and agreed to the published version of the manuscript.

Funding

We would like to acknowledge the support from the National Natural Science Foundation of China (Nos. 12374390, 32025021, and 52501219), the Noncommunicable Chronic Diseases–National Science and Technology Major Project (2023ZD0500902), the member of Youth Innovation Promotion Association Foundation of CAS, China (2023310), the Key Scientific and Technological Special Project of Ningbo City (2023Z209), and the Ningbo Youth Science and Technology Innovation Leading Talents Project (2024QL029), Ningbo Yongjiang Talent Programme (2025A-152-G).

Conflicts of Interest

The authors declare no conflict of interest.

Use of AI and AI-Assisted Technologies

No AI tools were utilized for this paper.

References

1. Bumbrah, G.S.; Sharma, R.M. Raman spectroscopy—Basic principle, instrumentation and selected applications for the characterization of drugs of abuse. *Egypt. J. Forensic Sci.* **2016**, *6*, 209–215.
2. Chandra, A.; Kumar, V.; Garnaik, U.C.; et al. Unveiling the Molecular Secrets: A Comprehensive Review of Raman Spectroscopy in Biological Research. *ACS Omega* **2024**, *9*, 50049–50063.
3. Yogurtecu, B.; Cebi, N.; Koçer, A.T.; et al. A Review of Non-Destructive Raman Spectroscopy and Chemometric Techniques in the Analysis of Cultural Heritage. *Molecules* **2024**, *29*, 5324.
4. Fernández-Galiana, Á.; Bibikova, O.; Vilms Pedersen, S.; et al. Fundamentals and Applications of Raman-Based Techniques for the Design and Development of Active Biomedical Materials. *Adv. Mater.* **2024**, *36*, 2210807.
5. Wachsmann-Hogiu, S.; Weeks, T.; Huser, T. Chemical analysis in vivo and in vitro by Raman spectroscopy—From single cells to humans. *Curr. Opin. Biotechnol.* **2009**, *20*, 63–73.
6. Albrecht, M.G.; Creighton, J.A. Anomalously intense Raman spectra of pyridine at a silver electrode. *J. Am. Chem. Soc.* **1977**, *99*, 5215–5217.
7. Pilot, R.; Signorini, R.; Durante, C.; et al. A Review on Surface-Enhanced Raman Scattering. *Biosensors* **2019**, *9*, 57.
8. Ding, S.-Y.; You, E.-M.; Tian, Z.-Q.; et al. Electromagnetic theories of surface-enhanced Raman spectroscopy. *Chem. Soc. Rev.* **2017**, *46*, 4042–4076.
9. Moskovits, M. Surface-enhanced spectroscopy. *Rev. Mod. Phys.* **1985**, *57*, 783–826.
10. Terzapulo, X.; Kassenova, A.; Bukasov, R. Immunoassays: Analytical and Clinical Performance, Challenges, and Perspectives of SERS Detection in Comparison with Fluorescent Spectroscopic Detection. *Int. J. Mol. Sci.* **2024**, *25*, 2080.
11. Tong, T.-T.; Xue, Y.-D.; Zhang, Y.-Y.; et al. Attenuation of arsenic trioxide-induced endothelial injury: Unveiling the protective role of ginkgolic acid through inhibition of TRPM4 SUMOylation: Evidence from Raman cellular imaging. *View* **2026**, *7*, 20250049.
12. Tang, Y.; Zheng, X.; Gao, T. Orthogonal Combinatorial Raman Codes Enable Rapid High-Throughput-Out Library Screening of Cell-Targeting Ligands. *Research* **2023**, *6*, 0136.
13. Bray, F.; Laversanne, M.; Sung, H.; et al. Global cancer statistics 2022: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA A Cancer J. Clin.* **2024**, *74*, 229–263.
14. Global, regional, and national burden of disorders affecting the nervous system, 1990-2021: A systematic analysis for the Global Burden of Disease Study 2021. *Lancet Neurol* **2024**, *23*, 344–381.
15. Lindstrom, M.; DeCleene, N.; Dorsey, H.; et al. Global Burden of Cardiovascular Diseases and Risks Collaboration, 1990-2021. *J. Am. Coll. Cardiol.* **2022**, *80*, 2372–2425.
16. Zhang, X.-H.; Wang, B.; Zhou, B.; et al. Recent advances in MXene-based flexible pressure sensors for medical monitoring. *Rare Met.* **2025**, *44*, 3653–3685.
17. Momenbeitollahi, N.; Cloet, T.; Li, H. Pushing the detection limits: Strategies towards highly sensitive optical-based protein detection. *Anal. Bioanal. Chem.* **2021**, *413*, 5995–6011.
18. Rusling, J.F.; Forster, R.J. Biosensors Designed for Clinical Applications. *Biomedicines* **2021**, *9*, 702.
19. Boschetti, E.; D'Amato, A.; Candiano, G.; et al. Protein biomarkers for early detection of diseases: The decisive contribution of combinatorial peptide ligand libraries. *J. Proteom.* **2018**, *188*, 1–14.
20. Zhang, C.; Li, Z.; Liu, J.; et al. Synthetic Gene Circuit-Based Assay with Multilevel Switch Enables Background-Free and Absolute Quantification of Circulating Tumor DNA. *Research* **2023**, *6*, 0217.
21. Li, L.-L.; Zhao, Y.; Pan, L.-J.; et al. A direct electrochemical biosensor for rapid glucose detection based on nitrogen-doped carbon nanocages. *Rare Met.* **2024**, *43*, 2184–2192.
22. Li, C.-z.; Hu, T.Y. Nanotechnology Powered CRISPR-Cas Systems for Point of Care Diagnosis and Therapeutic. *Research* **2022**, *2022*, 2.
23. Zhong, S.-J.; Chen, K.-Y.; Wang, S.-L.; et al. Metal-based nanowires in electrical biosensing. *Rare Met.* **2024**, *43*, 6233–6254.
24. Ding, Y.; Chen, J.; Wu, Q.; et al. Artificial intelligence-assisted point-of-care testing system for ultrafast and quantitative detection of drug-resistant bacteria. *SmartMat* **2024**, *5*, e1214.
25. Li, G.; Fan, X.; Tang, X.; et al. Challenges and Prospects of Personalized Healthcare Based on Surface-Enhanced Raman Spectroscopy. *Research* **2024**, *7*, 0572. <https://doi.org/10.34133/research.0572>.
26. Moore, T.J.; Moody, A.S.; Payne, T.D.; et al. In Vitro and In Vivo SERS Biosensing for Disease Diagnosis. *Research* **2018**, *8*, 46.
27. Elsheikh, S.; Coles, N.P.; Achadu, O.J.; et al. Advancing Brain Research through Surface-Enhanced Raman Spectroscopy (SERS): Current Applications and Future Prospects. *Biosensors* **2024**, *14*, 33.
28. Pollap, A.; Świt, P. Recent Advances in Sandwich SERS Immunosensors for Cancer Detection. *Int. J. Mol. Sci.* **2022**, *23*, 4740.
29. Liu, A.; Gao, C.; Xue, J.; et al. Toward reproducible SERS biosensing: Analysing instability origins and mitigation approaches from sample preparation to detection. *J. Mater. Chem. B* **2026**, *14*, 3071–3092.

30. Tas, Z.; Ciftci, F.; Icoz, K.; et al. Emerging biomedical applications of surface-enhanced Raman spectroscopy integrated with artificial intelligence and microfluidic technologies. *Spectrochim. Acta Part A Mol. Biomol. Spectrosc.* **2025**, *339*, 126285.
31. Ding, S.-Y.; Yi, J.; Li, J.-F.; et al. Nanostructure-based plasmon-enhanced Raman spectroscopy for surface analysis of materials. *Nat. Rev. Mater.* **2016**, *1*, 16021. <https://doi.org/10.1038/natrevmats.2016.21>.
32. Zhao, Z.-H.; Ma, S.-X.; Pan, Y.-F.; et al. Flexible and multifunctional humidity sensor based on Au@ZIF-67 nanoparticles for non-contact healthcare monitoring. *Rare Met.* **2025**, *44*, 2564–2576.
33. Stone, J.; Jackson, S.; Wright, D. Biological applications of gold nanorods. *Wiley Interdiscip. Rev. Nanomed. Nanobiotechnol.* **2011**, *3*, 100–109.
34. Mayer, K.M.; Hafner, J.H. Localized surface plasmon resonance sensors. *Chem. Rev.* **2011**, *111*, 3828–3857.
35. Kelly, K.L.; Coronado, E.; Zhao, L.L.; et al. The Optical Properties of Metal Nanoparticles: The Influence of Size, Shape, and Dielectric Environment. *J. Phys. Chem. B* **2003**, *107*, 668–677.
36. Jiang, X.; Qu, D.-Y.; Zhang, J.-N.; et al. Confronting pressure and humidity limitations in gold nanoparticle sensors array for enhancing electronic nose technology in real-world applications. *Rare Met.* **2025**, *44*, 8789–8801.
37. Tiwari, P.M.; Vig, K.; Dennis, V.A.; et al. Functionalized Gold Nanoparticles and Their Biomedical Applications. *Nanomaterials* **2011**, *1*, 31–63.
38. Zhang, J.; Mou, L.; Jiang, X. Surface chemistry of gold nanoparticles for health-related applications. *Chem. Sci.* **2020**, *11*, 923–936.
39. Amina, S.J.; Guo, B. A Review on the Synthesis and Functionalization of Gold Nanoparticles as a Drug Delivery Vehicle. *Int. J. Nanomed.* **2020**, *15*, 9823–9857.
40. Wan, H.; Ye, T.; Bi, Y.; et al. Nanobody–chlorin e6 conjugate for Nectin-4-mediated tumor targeting and enhanced photodynamic therapy. *View* **2025**, 20250185. <https://doi.org/10.1002/VIW.20250185>.
41. Alex, S.; Tiwari, A. Functionalized Gold Nanoparticles: Synthesis, Properties and Applications—A Review. *J. Nanosci. Nanotechnol.* **2015**, *15*, 1869–1894.
42. Li, M.; Wei, J.; Song, Y.; et al. Gold nanocrystals: Optical properties, fine-tuning of the shape, and biomedical applications. *RSC Adv.* **2022**, *12*, 23057–23073.
43. Hang, Y.; Wang, A.; Wu, N. Plasmonic silver and gold nanoparticles: Shape- and structure-modulated plasmonic functionality for point-of-care sensing, bio-imaging and medical therapy. *Chem. Soc. Rev.* **2024**, *53*, 2932–2971.
44. López-Lorente, Á.I. Recent developments on gold nanostructures for surface enhanced Raman spectroscopy: Particle shape, substrates and analytical applications. A review. *Anal. Chim. Acta* **2021**, *1168*, 338474.
45. Huang, X.; Neretina, S.; El-Sayed, M.A. Gold nanorods: From synthesis and properties to biological and biomedical applications. *Adv. Mater.* **2009**, *21*, 4880–4910.
46. Chen, C.; Zheng, L.; Guo, F.; et al. Programmable Self-Assembly of Gold Nanoarrows via Regioselective Adsorption. *Research* **2021**, *2021*, 10.
47. Das, G.M.; Managò, S.; Mangini, M.; et al. Biosensing Using SERS Active Gold Nanostructures. *Nanomater. (Basel)* **2021**, *11*, 2679.
48. Fan, M.; Andrade, G.F.S.; Brolo, A.G. A review on the fabrication of substrates for surface enhanced Raman spectroscopy and their applications in analytical chemistry. *Anal. Chim. Acta* **2011**, *693*, 7–25.
49. Awiaz, G.; Lin, J.; Wu, A. Recent advances of Au@Ag core-shell SERS-based biosensors. *Exploration* **2023**, *3*, 20220072.
50. Liu, R.; Liu, B.; Guan, G.; et al. Multilayered shell SERS nanotags with a highly uniform single-particle Raman readout for ultrasensitive immunoassays. *Chem. Commun.* **2012**, *48*, 9421–9423.
51. Fu, Q.; Liu, H.L.; Wu, Z.; et al. Rough surface Au@Ag core-shell nanoparticles to fabricating high sensitivity SERS immunochromatographic sensors. *J. Nanobiotechnol.* **2015**, *13*, 81.
52. Lu, W.; Zhou, F.; He, X.; et al. Smart polymeric hydrogels. *SmartMat* **2024**, *5*, e1282.
53. Lan, T.; Cui, D.; Liu, T.; et al. Gold NanoStars: Synthesis, Modification and Application. *Nano Biomed. Eng.* **2023**, *15*, 330–341. <https://doi.org/10.26599/NBE.2023.9290025>.
54. Reguera, J.; Langer, J.; Jiménez de Aberasturi, D.; et al. Anisotropic metal nanoparticles for surface enhanced Raman scattering. *Chem. Soc. Rev.* **2017**, *46*, 3866–3885.
55. Li, M.; Cushing, S.K.; Zhang, J.; et al. Shape-dependent surface-enhanced Raman scattering in gold-Raman probe-silica sandwiched nanoparticles for biocompatible applications. *Nanotechnology* **2012**, *23*, 115501.
56. Quang, A.T.N.; Nguyen, T.A.; Vu, S.V.; et al. Facile tuning of tip sharpness on gold nanostars by the controlled seed-growth method and coating with a silver shell for detection of thiram using surface enhanced Raman spectroscopy (SERS). *RSC Adv.* **2022**, *12*, 22815–22825.
57. Khosroshahi, M.E.; Patel, Y.; Chabok, R. Non-invasive optical characterization and detection of CA 15-3 breast cancer biomarker in blood serum using monoclonal antibody-conjugated gold nanourchin and surface-enhanced Raman scattering. *Lasers Med. Sci.* **2022**, *38*, 24.

58. Li, C.; Chen, P.; Wang, Z.; et al. A DNzyme-gold nanostar probe for SERS-fluorescence dual-mode detection and imaging of calcium ions in living cells. *Sens. Actuators B Chem.* **2021**, *347*, 130596.
59. Xiang, S.; Lu, L.; Zhong, H.; et al. SERS diagnosis of liver fibrosis in the early stage based on gold nanostar liver targeting tags. *Biomater. Sci.* **2021**, *9*, 5035–5044. <https://doi.org/10.1039/D1BM00013F>.
60. Cheng, R.; Tong Li, L.; Huang, M.; et al. Highly sensitive plasmonic biosensor for hepatitis B virus DNA based on the surface etching of the active helical gold nanorods. *Chem. Eng. J.* **2023**, *468*, 143627.
61. Dey, P.; Baumann, V.; Rodríguez-Fernández, J. Gold Nanorod Assemblies: The Roles of Hot-Spot Positioning and Anisotropy in Plasmon Coupling and SERS. *Nanomaterials* **2020**, *10*, 942.
62. Sharma, M.; Kaur, C.; Singhmar, P.; et al. DNA origami-templated gold nanorod dimer nanoantennas: Enabling addressable optical hotspots for single cancer biomarker SERS detection. *Nanoscale* **2024**, *16*, 15128–15140.
63. Su, A.; Liu, Y.; Cao, X.; et al. A universal CRISPR/Cas12a-mediated AuNPs aggregation-based surface-enhanced Raman scattering (CRISPR/Cas-SERS) platform for virus gene detection. *Sens. Actuators B Chem.* **2022**, *369*, 132295.
64. Wang, Y.; Liu, Y.; Li, Y.; et al. Magnetic Nanomotor-Based Maneuverable SERS Probe. *Research* **2020**, *2020*, 13.
65. Zhang, S.; Yin, G.; Wang, Z.; et al. Silicon-based Tubular Micromotor with SERS Traceability and Magnetic–Thermal Dual Responsiveness. *Nano Biomed. Eng.* **2025**, *17*, 263–276.
66. Tadi, S.R.; Shenoy, A.G.; Bharadwaj, A.; et al. Recent advances in the design of SERS substrates and sensing systems for (bio)sensing applications: Systems from single cell to single molecule detection. *F1000Research* **2024**, *13*, 670.
67. Bathini, S.; Raju, D.; Badilescu, S.; et al. Nano–Bio Interactions of Extracellular Vesicles with Gold Nanoislands for Early Cancer Diagnosis. *Research* **2018**, *2018*, 10.
68. Zhao, S.; Zhang, Y.; Wen, J.; et al. Nanopores-coupled SERS platforms on superslippery surfaces using Au&Ag alloy pillars for biomolecules enrichment and detection. *Rare Met.* **2025**, *44*, 10404–10417.
69. Ge, K.; Hu, Y.; Li, G. Recent Progress on Solid Substrates for Surface-Enhanced Raman Spectroscopy Analysis. *Biosensors* **2022**, *12*, 941.
70. Lin, C.; Li, Y.; Peng, Y.; et al. Recent development of surface-enhanced Raman scattering for biosensing. *J Nanobiotechnology* **2023**, *21*, 149.
71. Pal, A.; Anu Roshini, R.; Varma, M.M. De-wetted gold nanostructures for SERS-based sensing of static and dynamic targets. *Appl. Surf. Sci.* **2024**, *678*, 161096.
72. Carreón, R.V.; Rodríguez-Hernández, A.G.; Serrano de la Rosa, L.E.; et al. Mechanically Flexible, Large-Area Fabrication of Three-Dimensional Dendritic Au Films for Reproducible Surface-Enhanced Raman Scattering Detection of Nanoplastics. *ACS Sens.* **2025**, *10*, 1747–1755.
73. Guo, J.; Mu, Z.-M.; Yu, J.; et al. Hierarchically porous ZIF-67-based Au with enhanced electromagnetic, chemical, and mass-transfer properties for flexible gas–liquid SERS sensing. *Rare Met.* **2025**, *44*, 7672–7685.
74. Mo, Y.; Zhang, X.; Zou, K.; et al. Au Ordered Array Substrate for Rapid Detection and Precise Identification of Etomidate in E-Liquid Through Surface-Enhanced Raman Spectroscopy. *Nanomaterials* **2024**, *14*, 1958.
75. Lu, Y.; Lei, B.; Zhao, Q.; et al. Solid-State Au Nanocone Arrays Substrate for Reliable SERS Profiling of Serum for Disease Diagnosis. *ACS Omega* **2023**, *8*, 29836–29846.
76. Tan, H.-S.; Wang, T.; Han, J.-M.; et al. Dual-signal SERS biosensor based on spindle-shaped gold array for sensitive and accurate detection of miRNA 21. *Sens. Actuators B Chem.* **2024**, *403*, 135157.
77. Gao, T.; Yachi, T.; Shi, X.; et al. Ultrasensitive Surface-Enhanced Raman Scattering Platform for Protein Detection via Active Delivery to Nanogaps as a Hotspot. *ACS Nano* **2024**, *18*, 21593–21606.
78. Gao, S.; Guo, Z.; Liu, Z. Recent Advances in Rational Design and Engineering of Signal-Amplifying Substrates for Surface-Enhanced Raman Scattering-Based Bioassays. *Chemosensors* **2023**, *11*, 461.
79. Li, J.F.; Huang, Y.F.; Ding, Y.; et al. Shell-isolated nanoparticle-enhanced Raman spectroscopy. *Nature* **2010**, *464*, 392–395.
80. Chang, J.; Zhang, A.; Huang, Z.; et al. Monodisperse Au@Ag core-shell nanoprobe with ultrasensitive SERS-activity for rapid identification and Raman imaging of living cancer cells. *Talanta* **2019**, *198*, 45–54.
81. Khlebtsov, N.G.; Lin, L.; Khlebtsov, B.N.; et al. Gap-enhanced Raman tags: Fabrication, optical properties, and theranostic applications. *Theranostics* **2020**, *10*, 2067–2094.
82. Wang, D.; Zhao, Y.; Zhang, S.; et al. Reporter Molecules Embedded Au@Ag Core-Shell Nanospheres as SERS Nanotags for Cardiac Troponin I Detection. *Biosensors* **2022**, *12*, 1108.
83. Khlebtsov, B.; Pylaev, T.; Khanadeev, V.; et al. Quantitative and multiplex dot-immunoassay using gap-enhanced Raman tags. *RSC Adv.* **2017**, *7*, 40834–40841.
84. Huang, Y.; Lin, D.; Li, M.; et al. Ag@Au Core–Shell Porous Nanocages with Outstanding SERS Activity for Highly Sensitive SERS Immunoassay. *Sensors* **2019**, *19*, 1554.
85. Yang, L.; Gao, M.X.; Zhan, L.; et al. An enzyme-induced Au@Ag core-shell nanostructure used for an ultrasensitive surface-enhanced Raman scattering immunoassay of cancer biomarkers. *Nanoscale* **2017**, *9*, 2640–2645.

86. Ning, C.-F.; Wang, L.; Tian, Y.-F.; et al. Multiple and sensitive SERS detection of cancer-related exosomes based on gold–silver bimetallic nanotrepangs. *Analyst* **2020**, *145*, 2795–2804.
87. Laing, S.; Jamieson, L.E.; Faulds, K.; et al. Surface-enhanced Raman spectroscopy for in vivo biosensing. *Nat. Rev. Chem.* **2017**, *1*, 0060. <https://doi.org/10.1038/s41570-017-0060>.
88. Gangwar, R.K.; Pathak, A.K.; Chiavaioli, F.; et al. Optical fiber SERS sensors: Unveiling advances, challenges, and applications in a miniaturized technology. *Coord. Chem. Rev.* **2024**, *510*, 215861. <https://doi.org/10.1016/j.ccr.2024.215861>.
89. Bratash, O.; Buhot, A.; Leroy, L.; et al. Optical fiber biosensors toward in vivo detection. *Biosens. Bioelectron.* **2024**, *251*, 116088. <https://doi.org/10.1016/j.bios.2024.116088>.
90. Ran, Y.; Strobba, P.; Cupil-Garcia, V.; et al. Fiber-optrode SERS probes using plasmonic silver-coated gold nanostars. *Sens. Actuators B Chem.* **2019**, *287*, 95–101. <https://doi.org/10.1016/j.snb.2019.01.167>.
91. Pisco, M.; Quero, G.; Cutolo, M.A.; et al. Lab-on-fiber technology: Towards engineered SERS optrodes. *APL Photonics* **2026**, *11*, 040901. <https://doi.org/10.1063/5.0307589>.
92. Spaziani, S.; Quero, G.; Managò, S.; et al. SERS assisted sandwich immunoassay platforms for ultrasensitive and selective detection of human Thyroglobulin. *Biosens. Bioelectron.* **2023**, *233*, 115322. <https://doi.org/10.1016/j.bios.2023.115322>.
93. Gao, D.; Liu, J.; Liu, X.; et al. Emerging frontiers in SERS-integrated optical waveguides: Advancing portable and ultra-sensitive detection for trace liquid analysis. *Light Sci. Appl.* **2025**, *14*, 389. <https://doi.org/10.1038/s41377-025-01989-6>.
94. Cong, S.; Liu, X.; Jiang, Y.; et al. Surface Enhanced Raman Scattering Revealed by Interfacial Charge-Transfer Transitions. *Innovation* **2020**, *1*, 100051. <https://doi.org/10.1016/j.xinn.2020.100051>.
95. Liang, X.; Li, N.; Zhang, R.; et al. Carbon-based SERS biosensor: From substrate design to sensing and bioapplication. *NPG Asia Mater.* **2021**, *13*, 8.
96. Meng, X.-Y.; Xie, Y.-J.; Sun, L.; et al. Electromagnetic field-charge transfer synergy boosted SERS in noble metal–semiconductor nanohybrids for environmental and bio-detection. *Rare Met.* **2025**, *44*, 9702–9725.
97. Song, X.; Xu, L.; Meng, X.; et al. TMO-based SERS: Dual enhancement mechanisms and multi-functional analytical applications. *Chin. J. Struct. Chem.* **2026**, *45*, 100797. <https://doi.org/10.1016/j.cjsc.2025.100797>.
98. Zhang, M.; Liu, A.; Zhang, J.; et al. Chemical Engineering of Heterojunction SERS Substrates: Emerging Tools for Disease Diagnosis and Health Monitoring. *SmartMat* **2025**, *6*, e70045. <https://doi.org/10.1002/smm2.70045>.
99. Li, M.-N.; Cao, B.; Wang, Y.-L.; et al. V2CTx MXene-powered handheld SERS biosensor for the viral antigen test. *Rare Met.* **2025**, *44*, 6442–6455.
100. Song, X.; Liu, X.; Xu, L.; et al. Sub-nanoscale oxygen-defect-rich MoO_{3-x}: A versatile platform for label-free ultrasensitive SERS biodetection. *Chin. J. Struct. Chem.* **2026**, *45*, 100848. <https://doi.org/10.1016/j.cjsc.2025.100848>.
101. Guo, L.; Mao, Z.; Jin, S.; et al. A SERS Study of Charge Transfer Process in Au Nanorod-MBA@Cu(2)O Assemblies: Effect of Length to Diameter Ratio of Au Nanorods. *Nanomaterials* **2021**, *11*, 867.
102. Zhang, M.; Meng, X.; Yu, J.; et al. A novel Fe₂O₃@CeO₂ heterojunction substrate with high surface-enhanced Raman scattering performance. *SmartMat* **2024**, *5*, e1301.
103. Zhao, S.; Zhao, Y.; Tao, L. Interface engineering in 2D materials for SERS sensing. *Front. Mater.* **2023**, *10*, 1272826.
104. Wang, Q.; Chang, K.; Yang, Q.; et al. Semiconductor-based surface-enhanced Raman scattering sensing platforms: State of the art, applications and prospects in food safety. *Trends Food Sci. Technol.* **2024**, *147*, 104460.
105. Lin, J.; Meng, X.; Xie, Y.; et al. Selectivity and stability reshaping high-sensitivity detection boundaries: A technical leap and paradigm shift in semiconductor surface-enhanced Raman scattering. *Nano Res.* **2026**, *19*, 94908347. <https://doi.org/10.26599/NR.2026.94908347>.
106. Huang, Z.-H.; Peng, L.-X.; Liu, X.-L.; et al. Advancing sensing frontiers: Elevating performance metrics and extending applications through two-dimensional materials. *Rare Met.* **2025**, *44*, 721–756.
107. Peng, Y.; Zhang, W.; Xu, M.; et al. Morphology Engineering of SnS₂ Nanostructures to Stimulate PICT Resonance for Ultra-Sensitive SERS Sensors. *Exploration* **2025**, *5*, 270016.
108. Adesoye, S.; Dellinger, K. ZnO and TiO₂ nanostructures for surface-enhanced Raman scattering-based bio-sensing: A review. *Sens. Bio-Sens. Res.* **2022**, *37*, 100499.
109. Yin, Y.; Li, C.; Yan, Y.; et al. MoS₂-Based Substrates for Surface-Enhanced Raman Scattering: Fundamentals, Progress and Perspective. *Coatings* **2022**, *12*, 360.
110. Liu, L.; Yang, H.; Ren, X.; et al. Au–ZnO hybrid nanoparticles exhibiting strong charge-transfer-induced SERS for recyclable SERS-active substrates. *Nanoscale* **2015**, *7*, 5147–5151.
111. Yang, L.; Ruan, W.; Jiang, X.; et al. Contribution of ZnO to Charge-Transfer Induced Surface-Enhanced Raman Scattering in Au/ZnO/PATP Assembly. *J. Phys. Chem. C* **2009**, *113*, 117–120.
112. Yu, H.; Sun, H.; Ma, J.; et al. Resonance-Assisted Surface-Enhanced Raman Spectroscopy Amplification on Hierarchical Rose-Shaped MoS₂/Au Nanocomposites. *Langmuir* **2024**, *40*, 380–388.
113. Li, J.; Xu, J.; Liu, Y.; et al. Hydrophobic interaction enables rapid enrichment of volatile metabolites on Au/TiO₂ based SERS substrates for ultrasensitive bacteria detection. *J. Mater. Chem. B* **2023**, *11*, 3877–3884.

114. Wei, Q.; Dong, Q.; Sun, D.-W.; et al. Synthesis of recyclable SERS platform based on MoS₂@TiO₂@Au heterojunction for photodegradation and identification of fungicides. *Spectrochim. Acta Part A Mol. Biomol. Spectrosc.* **2023**, *285*, 121895.
115. Qian, W.; Xing, M.; Ye, M.; et al. Reproducible and acid-responsive Rhodamine B/PEG functionalized nanographene oxide-Au nanocomposites for surface-enhanced Raman scattering sensing. *SmartMat* **2024**, *5*, e1305.
116. Banihashemi Jozdani, S.M.; Hashemian, Z.; Ebrahim Damavandi, S.; et al. Emerging Trends in the Biomedical Application of Carbon-based Nanomaterials. *Nano Biomed. Eng.* **2024**, *16*, 357–369.
117. Hernández-Sánchez, D.; Villabona-Leal, G.; Saucedo-Orozco, I.; et al. Stable graphene oxide-gold nanoparticle platforms for biosensing applications. *Phys. Chem. Chem. Phys.* **2018**, *20*, 1685–1692.
118. Lee, J.; Kim, J.; Kim, S.; et al. Biosensors based on graphene oxide and its biomedical application. *Adv. Drug Deliv. Rev.* **2016**, *105*, 275–287.
119. Zhu, X.; Shi, L.; Schmidt, M.S.; et al. Enhanced Light–Matter Interactions in Graphene-Covered Gold Nanovoid Arrays. *Nano Lett.* **2013**, *13*, 4690–4696.
120. Pan, X.; Li, L.; Lin, H.; et al. A graphene oxide-gold nanostar hybrid based-paper biosensor for label-free SERS detection of serum bilirubin for diagnosis of jaundice. *Biosens. Bioelectron.* **2019**, *145*, 111713.
121. Leem, J.; Wang, M.C.; Kang, P.; et al. Mechanically Self-Assembled, Three-Dimensional Graphene–Gold Hybrid Nanostructures for Advanced Nanoplasmonic Sensors. *Nano Lett.* **2015**, *15*, 7684–7690.
122. He, J.; Hou, Y.; Zhang, Z.; et al. Carbon-based Nanozymes: How Structure Affects Performance. *Nano Biomed. Eng.* **2024**, *16*, 28–47. <https://doi.org/10.26599/NBE.2024.9290053>.
123. Qian, H.; Zhu, M.; Wu, Z.; et al. Quantum sized gold nanoclusters with atomic precision. *Acc. Chem. Res.* **2012**, *45*, 1470–1479.
124. Aikens, C.M. Electronic and Geometric Structure, Optical Properties, and Excited State Behavior in Atomically Precise Thiolate-Stabilized Noble Metal Nanoclusters. *Acc. Chem. Res.* **2018**, *51*, 3065–3073.
125. Shang, L.; Nienhaus, G.U. Gold nanoclusters as novel optical probes for in vitro and in vivo fluorescence imaging. *Biophys. Rev.* **2012**, *4*, 313–322.
126. Risse, T.; Shaikhutdinov, S.; Nilius, N.; et al. Gold supported on thin oxide films: From single atoms to nanoparticles. *Acc. Chem. Res.* **2008**, *41*, 949–956.
127. Sun, L.; Zhao, S.; Zheng, S.; et al. Recent advances of metal cluster-based SERS probes for biomedical applications. *Nanoscale* **2025**, *17*, 13998–14015. <https://doi.org/10.1039/d5nr01131k>.
128. Wang, Y.; Meng, X.; Shi, W.; et al. Single-Atom Cu Anchored on a UiO-66 Surface-Enhanced Raman Scattering Sensor for Trace and Rapid Detection of Volatile Organic Compounds. *Research* **2025**, *8*, 0841.
129. Hu, M.-E.; Wang, Z.; Gao, X.-C.; et al. Tantalum-doped tungsten carbide nanostructures with efficient charge transfer for superior SERS performance. *Rare Met.* **2025**, *44*, 8802–8812.
130. You, T.; Liang, X.; Gao, Y.; et al. A computational study on surface-enhanced Raman spectroscopy of para-substituted Benzenethiol derivatives adsorbed on gold nanoclusters. *Spectrochim. Acta Part A Mol. Biomol. Spectrosc.* **2016**, *152*, 278–287.
131. Boto, R.A.; Esteban, R.; Candelas, B.; et al. Theoretical Procedure for Precise Evaluation of Chemical Enhancement in Molecular Surface-Enhanced Raman Scattering. *J. Phys. Chem. C* **2024**, *128*, 18293–18304.
132. Yu, J.; Chen, C.; Zhang, Q.; et al. Au Atoms Anchored on Amorphous C(3)N(4) for Single-Site Raman Enhancement. *J Am Chem Soc* **2022**, *144*, 21908–21915.
133. Wen, C.; Wang, L.; Liu, L.; et al. Surface-Enhanced Raman Probes Based on Gold Nanomaterials for in vivo Diagnosis and Imaging. *Chem. Asian J.* **2022**, *17*, e202200014.
134. Cialla-May, D.; Bonifacio, A.; Bocklitz, T.; et al. Biomedical SERS - the current state and future trends. *Chem. Soc. Rev.* **2024**, *53*, 8957–8979.
135. Lyu, N.; Hassanzadeh-Barforoushi, A.; Rey Gomez, L.M.; et al. SERS biosensors for liquid biopsy towards cancer diagnosis by detection of various circulating biomarkers: Current progress and perspectives. *Nano Converg* **2024**, *11*, 22.
136. Sun, Y.; Shi, L.; Mi, L.; et al. Recent progress of SERS optical nanosensors for miRNA analysis. *J. Mater. Chem. B* **2020**, *8*, 5178–5183. <https://doi.org/10.1039/D0TB00280A>.
137. Wu, W.; Ali, A.; Sun, L.; et al. Nanobodies: A Promising Toolkit for Diagnostic Applications. *SmartMat* **2026**, *7*, e70088. <https://doi.org/10.1002/smm2.70088>.
138. Tavakkoli Yarak, M.; Tukova, A.; Wang, Y. Emerging SERS biosensors for the analysis of cells and extracellular vesicles. *Nanoscale* **2022**, *14*, 15242–15268. <https://doi.org/10.1039/D2NR03005E>.
139. Dell’Olio, F. Multiplexed Liquid Biopsy and Tumor Imaging Using Surface-Enhanced Raman Scattering. *Biosensors* **2021**, *11*, 449.
140. Tan, E.X.; Zhong, Q.Z.; Ting Chen, J.R.; et al. Surface-Enhanced Raman Scattering-Based Multimodal Techniques: Advances and Perspectives. *ACS Nano* **2024**, *18*, 32315–32334. <https://doi.org/10.1021/acsnano.4c12996>.

141. Mensah, G.A.; Fuster, V.; Murray, C.J.L.; et al. Global Burden of Cardiovascular Diseases and Risks, 1990–2022. *J. Am. Coll. Cardiol.* **2023**, *82*, 2350–2473.
142. Chang, X.; Wang, H.; Chen, X. Tumor Diagnosis and Treatment Based on Stimuli-Responsive Aggregation of Gold Nanoparticles. *Exploration* **2025**, *5*, 270006.
143. Miao, X.; Xu, L.; Sun, L.; et al. Highly Sensitive Detection and Molecular Subtyping of Breast Cancer Cells Using Machine Learning-assisted SERS Technology. *Nano Biomed. Eng.* **2025**, *17*, 129–142. <https://doi.org/10.26599/nbe.2025.9290113>.
144. Li, N.; Bian, F.; Wei, X.; et al. Pollen-Inspired Photonic Barcodes with Prickly Surface for Multiplex Exosome Capturing and Screening. *Research* **2022**, *2022*, 9.
145. Guo, W.; Cai, Y.; Liu, X.; et al. Single-Exosome Profiling Identifies ITGB3+ and ITGAM+ Exosome Subpopulations as Promising Early Diagnostic Biomarkers and Therapeutic Targets for Colorectal Cancer. *Research* **2023**, *6*, 0041.
146. Hanjani, N.A.; Esmaelizad, N.; Zanganeh, S.; et al. Emerging role of exosomes as biomarkers in cancer treatment and diagnosis. *Crit. Rev. Oncol. /Hematol.* **2022**, *169*, 103565.
147. Metcalf, G.A.D. MicroRNAs: Circulating biomarkers for the early detection of imperceptible cancers via biosensor and machine-learning advances. *Oncogene* **2024**, *43*, 2135–2142.
148. Guo, Y.; Zhang, R.; You, H.; et al. Effective enrichment of trace exosomes for the label-free SERS detection via low-cost thermophoretic profiling. *Biosens. Bioelectron.* **2024**, *253*, 116164.
149. Kang, T.; Zhu, J.; Luo, X.; et al. Controlled Self-Assembly of a Close-Packed Gold Octahedra Array for SERS Sensing Exosomal MicroRNAs. *Anal. Chem.* **2021**, *93*, 2519–2526.
150. Si, Y.; Xu, L.; Deng, T.; et al. Catalytic Hairpin Self-Assembly-Based SERS Sensor Array for the Simultaneous Measurement of Multiple Cancer-Associated miRNAs. *ACS Sens.* **2020**, *5*, 4009–4016.
151. Li, J.Q.; Neng-Wang, H.; Canning, A.J.; et al. Surface-Enhanced Raman Spectroscopy-Based Detection of Micro-RNA Biomarkers for Biomedical Diagnosis Using a Comparative Study of Interpretable Machine Learning Algorithms. *Appl. Spectrosc.* **2024**, *78*, 84–98.
152. Xu, L.; Xie, Y.; Lin, J.; et al. Advancements in SERS-based biological detection and its application and perspectives in pancreatic cancer. *View* **2024**, *5*, 20230070. <https://doi.org/10.1002/VIW.20230070>.
153. Zhu, S.; Zhu, Z.; Ni, C.; et al. Liquid Biopsy Instrument for Ultra-Fast and Label-Free Detection of Circulating Tumor Cells. *Research* **2024**, *7*, 0431.
154. Linkner, T.R.; Nagy, Z.B.; Kalmár, A.; et al. Circulating tumor cells: Indicators of cancer progression, plasticity and utility for therapies. *Pathol. Oncol. Res.* **2025**, *31*, 1612181.
155. Allen, T.A. The Role of Circulating Tumor Cells as a Liquid Biopsy for Cancer: Advances, Biology, Technical Challenges, and Clinical Relevance. *Cancers* **2024**, *16*, 1377.
156. Sha, M.Y.; Xu, H.; Natan, M.J.; et al. Surface-Enhanced Raman Scattering Tags for Rapid and Homogeneous Detection of Circulating Tumor Cells in the Presence of Human Whole Blood. *J. Am. Chem. Soc.* **2008**, *130*, 17214–17215.
157. Miao, X.; Zhang, Z.; Cai, X.; et al. Ratiometric SERS platform of dual bioprobes for stable detection of circulating tumor cells. *Nano Biomed. Eng.* **2026**, *18*, 100019. <https://doi.org/10.1016/j.nbe.2025.100019>.
158. Bhana, S.; Wang, Y.; Huang, X. Nanotechnology for enrichment and detection of circulating tumor cells. *Nanomedicine* **2015**, *10*, 1973–1990.
159. Xue, T.; Wang, S.; Ou, G.; et al. Detection of circulating tumor cells based on improved SERS-active magnetic nanoparticles. *Anal. Methods* **2019**, *11*, 2918–2928.
160. Tang, R.; Hu, R.; Jiang, X.; et al. LHRH-targeting surface-enhanced Raman scattering tags for the rapid detection of circulating tumor cells. *Sens. Actuators B Chem.* **2019**, *284*, 468–474.
161. Wu, X.; Xia, Y.; Huang, Y.; et al. Improved SERS-Active Nanoparticles with Various Shapes for CTC Detection without Enrichment Process with Supersensitivity and High Specificity. *ACS Appl. Mater. Interfaces* **2016**, *8*, 19928–19938.
162. Wang, J.; Zhang, R.; Ji, X.; et al. SERS and fluorescence detection of circulating tumor cells (CTCs) with specific capture-release mode based on multifunctional gold nanomaterials and dual-selective recognition. *Anal. Chim. Acta* **2021**, *1141*, 206–213.
163. Wang, Y.; Kang, S.; Khan, A.; et al. Quantitative molecular phenotyping with topically applied SERS nanoparticles for intraoperative guidance of breast cancer lumpectomy. *Sci. Rep.* **2016**, *6*, 21242.
164. Wang, Y.W.; Doerksen, J.D.; Kang, S.; et al. Multiplexed Molecular Imaging of Fresh Tissue Surfaces Enabled by Convection-Enhanced Topical Staining with SERS-Coded Nanoparticles. *Small* **2016**, *12*, 5612–5621.
165. Czaja, A.; Jiang, A.J.; Blanco, M.Z.; et al. A Raman topography imaging method toward assisting surgical tumor resection. *Npj Imaging* **2024**, *2*, 2.
166. Chang, J.; Guo, B.; Gao, Y.; et al. Characteristic Features of Deep Brain Lymphatic Vessels and Their Regulation by Chronic Stress. *Research* **2023**, *6*, 0120.

167. Zhao, Y.; Xiong, C.; Wang, B.; et al. The Discovery of Phages in the Substantia Nigra and Its Implication for Parkinson's Disease. *Research* **2025**, *8*, 0657.
168. Ni, R.; Rominger, A. Noninvasive imaging of amyloid-beta and tau in rodent and nonhuman primate models. *View* **2026**, *7*, 20250071.
169. Blennow, K.; Zetterberg, H. Fluid biomarker-based molecular phenotyping of Alzheimer's disease patients in research and clinical settings. *Prog. Mol. Biol. Transl. Sci.* **2019**, *168*, 3–23.
170. Pais, M.V.; Forlenza, O.V.; Diniz, B.S. Plasma Biomarkers of Alzheimer's Disease: A Review of Available Assays, Recent Developments, and Implications for Clinical Practice. *J. Alzheimer's Dis. Rep.* **2023**, *7*, 355–380.
171. Pradeepkiran, J.A.; Baig, J.; Islam, M.A.; et al. Amyloid- β and Phosphorylated Tau are the Key Biomarkers and Predictors of Alzheimer's Disease. *Aging Dis.* **2024**, *16*, 658–682.
172. Agnello, L.; Gambino, C.M.; Ciaccio, A.M.; et al. Molecular Biomarkers of Neurodegenerative Disorders: A Practical Guide to Their Appropriate Use and Interpretation in Clinical Practice. *Int. J. Mol. Sci.* **2024**, *25*, 4323.
173. Ibanez, L.; Liu, M.; Beric, A.; et al. Benchmarking of a multi-biomarker low-volume panel for Alzheimer's Disease and related dementia research. *medRxiv* **2024**, *21*, e14413.
174. Shim, J.-E.; Kim, Y.J.; Hahm, E.; et al. Ultrasensitive SERS nanoprobe-based multiplexed digital sensing platform for the simultaneous quantification of Alzheimer's disease biomarkers. *Biosens. Bioelectron.* **2025**, *274*, 117216.
175. Muhammad, M.; Liu, C.; Yang, G.; et al. Early-stage Alzheimer's disease profiling in blood achieved by multiplexing aptamer-SERS biosensors. *Biosens. Bioelectron.* **2025**, *268*, 116907.
176. Cehlar, O.; Njemoga, S.; Horvath, M.; et al. Structures of Oligomeric States of Tau Protein, Amyloid- β , α -Synuclein and Prion Protein Implicated in Alzheimer's Disease, Parkinson's Disease and Prionopathies. *Int. J. Mol. Sci.* **2024**, *25*, 13049.
177. Elbassal, E.A.; Morris, C.; Kent, T.W.; et al. Gold Nanoparticles as a Probe for Amyloid- β Oligomer and Amyloid Formation. *J. Phys. Chem. C* **2017**, *121*, 20007–20015.
178. Wang, L.; Chen, H.; Ma, S.; et al. Ultra-sensitive SERS detection of A β 1–42 for Alzheimer's disease using graphene oxide/gold nanohybrids. *Vib. Spectrosc.* **2023**, *129*, 103614.
179. Ho, W.K.H.; Zhang, Q.; Zhorabe, F.; et al. A buoyant plasmonic microbubble-based SERS sensing platform for amyloid-beta protein detection in Alzheimer's disease. *J. Mater. Chem. B* **2025**, *13*, 8883–8896.
180. Liu, M.; Li, M.; He, J.; et al. Chiral Amino Acid Profiling in Serum Reveals Potential Biomarkers for Alzheimer's Disease. *J. Alzheimer's Dis.* **2023**, *94*, 291–301.
181. Liu, Y.; Wu, Z.; Armstrong, D.W.; et al. Detection and analysis of chiral molecules as disease biomarkers. *Nat. Rev. Chem.* **2023**, *7*, 355–373.
182. Hao, C.; Meng, D.; Shi, W.; et al. Chiral Gold Nanostructure Monolayers as SERS Substrates for Ultrasensitive Detection of Enantiomer Biomarkers of Alzheimer's Disease. *Angew. Chem. Int. Ed.* **2025**, *64*, e202502115.
183. Ouyang, Y.-C.; Yeom, B.-J.; Zhao, Y.; et al. Progress and prospects of chiral nanomaterials for biosensing platforms. *Rare Met.* **2024**, *43*, 2469–2497.
184. Liu, H.; Ou, D.; Zhao, H.; et al. Immunological landscape of radiation-induced cardiac injury. *View* **2025**, *6*, 20250012.
185. Deng, X.; Wang, J.; Yu, S.; et al. Advances in the treatment of atherosclerosis with ligand-modified nanocarriers. *Exploration* **2024**, *4*, 20230090.
186. Lu, C.; Li, C.; Gu, N.; et al. Emerging Elastic Micro-Nano Materials for Diagnosis and Treatment of Thrombosis. *Research* **2025**, *8*, 0614.
187. Davronova, G.; Siradjitdinova, N.; Munira, K.; et al. Optical nanobiosensing of cardiovascular disease. *Clin. Chim. Acta* **2026**, *582*, 120800.
188. Yoo, B.S. Clinical Significance of B-type Natriuretic Peptide in Heart Failure. *J. Lifestyle Med.* **2014**, *4*, 34–38.
189. Wang, X.Y.; Zhang, F.; Zhang, C.; et al. The Biomarkers for Acute Myocardial Infarction and Heart Failure. *Biomed. Res. Int.* **2020**, *2020*, 2018035.
190. Xiang, Q.; Wang, H.; Liu, S.; et al. Highly sensitive and reproducible SERS substrate based on ordered multi-tipped Au nanostar arrays for the detection of myocardial infarction biomarker cardiac troponin I. *Analyst* **2025**, *150*, 2239–2250.
191. Lin, C.; Li, L.; Feng, J.; et al. Aptamer-modified magnetic SERS substrate for label-based determination of cardiac troponin I. *Microchim. Acta* **2021**, *189*, 22.
192. Lim, W.Y.; Goh, C.H.; Thevarajah, T.M.; et al. Using SERS-based microfluidic paper-based device (μ PAD) for calibration-free quantitative measurement of AMI cardiac biomarkers. *Biosens Bioelectron.* **2020**, *147*, 111792.
193. Cai, P.; Xu, T.; Wei, H.; et al. Compact All-Fiber SERS Probe Sensor Based on the MMF-NCF Structure with Self-Assembled Gold Nanoparticles. *Sensors* **2025**, *25*, 5221.
194. Dong, P.; Xiong, S.; Gong, Y.; et al. SERS-based pump-free microfluidic chip sensor for immunoassays of the myocardial injury marker CK-MB. *Sens. Actuators B Chem.* **2026**, *451*, 139397.
195. Campu, A.; Muresan, I.; Potara, M.; et al. Portable microfluidic plasmonic chip for fast real-time cardiac troponin I biomarker thermoplasmonic detection. *J. Mater. Chem. B* **2024**, *12*, 962–972.

196. Dugandžić, V.; Drikermann, D.; Ryabchykov, O.; et al. Surface enhanced Raman spectroscopy-detection of the uptake of mannose-modified nanoparticles by macrophages in vitro: A model for detection of vulnerable atherosclerotic plaques. *J. Biophotonics* **2018**, *11*, e201800013.
197. Noonan, J.; Asiala, S.M.; Grassia, G.; et al. In vivo multiplex molecular imaging of vascular inflammation using surface-enhanced Raman spectroscopy. *Theranostics* **2018**, *8*, 6195–6209.