

A SERS Detection Platform Based on a Photocoupling Signal Transduction Mechanism for Highly Sensitive Detection of Serum SA- β -gal in Colitis

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Abstract

Colitis is an inflammatory bowel disease with an incidence rate that has been rising year by year; however, there is currently a lack of simple and sensitive serological tests for its early diagnosis. This study developed a surface-enhanced Raman scattering (SERS) detection platform based on a magnetic core-shell composite structure (Fe_3O_4 NRs@MPBA@ β -lactose-Au NPs@PATP/MPBA). This platform utilized a target-induced enzymatic release process to trigger a light-coupled signal amplification reaction, thereby enabling the sensitive detection of senescence-associated β -galactosidase (SA- β -gal), a serum biomarker of colorectal inflammation. During the detection process, the target SA- β -gal specifically hydrolyzed the galactosidic bonds in β -lactose, causing the SERS probe to dissociate and be released from the surface of the capture probe. Subsequently, p-aminobenzenethiol (PATP) on the surface of SERS probes could undergo an in situ photocoupling reaction upon laser irradiation, converting to 4,4'-dithioazobenzene (DMAB), as evidenced by a significant decrease in the characteristic Raman signal at 1397 cm^{-1} . The detection range of this SERS detection platform spanned 10^{-4} to 10^0 U/mL, with a limit of detection (LOD) as low as 1.956×10^{-6} U/mL. The platform showed good sensitivity and specificity in serum samples, and could effectively distinguish the difference of serum SA- β -gal levels between colitis model mice and healthy control mice. Compared with the colorimetric method, the detection results of actual serum samples had good accuracy. This study shows great promise for clinical application in the early serological screening of colorectal inflammation.

1. Introduction

Colitis is a disease with rectal and colonic mucosal inflammation as the core performance[1, 2]. According to the etiology, it can be divided into ulcerative, infectious, ischemic and radioactive colitis types[3, 4]. The core pathological features are mucosal epithelial barrier destruction, intestinal flora imbalance and abnormal activation of immune cells[5]. The clinical manifestations are recurrent abdominal pain, diarrhea, mucus purulent and bloody stool, and severe cases can be complicated with intestinal perforation and even canceration[6]. At present, the diagnosis of colitis mainly depends on endoscopy and histopathology[7]. However, this method has limitations such as strong invasion and poor patient compliance[8]. Fecal examination only plays a preliminary role in screening, but it has deficiencies in specificity and sensitivity, and it is unable to assess the scope of the disease and determine the cause[9, 10]. Therefore, finding a non-invasive and highly sensitive serum biomarker for colitis has important clinical value for early diagnosis and real-time monitoring of the disease.

Senescence-associated β -galactosidase (SA- β -gal) is widely distributed in the gut microbiota and belongs to the glycosidase family[11]. Its classic physiological function is to catalyze the breakdown of lactose into glucose and galactose, which can be easily absorbed by the intestine[12]. In cases of colitis, persistent inflammatory responses can damage the structure of the intestinal mucosa, leading to a significant reduction in SA- β -gal activity[13, 14]. Recent studies have shown that SA- β -gal, as an important biological marker reflecting the state of the intestinal mucosal barrier, plays a key role in regulating the homeostasis of the intestinal microenvironment[15]. Using a radiation-induced intestinal

injury model, Yang's research team further confirmed that the aging process of intestinal epithelial cells leads to a significant increase in SA- β -gal activity, accompanied by damage to the mucosal barrier and massive secretion of pro-inflammatory mediators[16]. However, existing SA- β -gal detection methods (such as colorimetric, fluorescent, and immunoassay methods) generally suffer from limited sensitivity and cumbersome procedures, making it difficult to meet the needs of early clinical diagnosis[17, 18]. Therefore, there is an urgent need to develop a simple, rapid, and ultra-sensitive detection method to screen and identify the risk of colorectal inflammation in large populations.

Surface-enhanced Raman scattering (SERS) is a powerful optical sensing technology capable of reflecting subtle biochemical changes in tissues at the molecular level[19–21]. It is minimally invasive, rapid, and offers “fingerprint”-like resolution[22, 23]. SERS technology utilizes the localized surface plasmon resonance (LSPR) effect generated by noble metal nanomaterials such as gold and silver to significantly amplify the inherently weak Raman signal, enabling ultra-sensitive detection of trace biomarkers[24, 25]. Compared to traditional detection methods, SERS technology is not subject to interference from aqueous solution signals, making it highly suitable for the analysis of clinical biological samples[26, 27]. Iron (III) oxide nanorods (Fe_3O_4 NRs), due to their unique tip-enhancement effect and excellent magnetic separation capabilities, hold broad application potential in the fields of biological sample analysis and disease biomarker detection[28, 29]. Compared to traditional spherical Fe_3O_4 nanomaterials, the rod-like structure possesses a higher specific surface area, a more pronounced tip-enhancement effect, and stronger magnetic enrichment capabilities, making it an ideal substrate material for constructing high-performance SERS biosensing platforms[30, 31].

In this study, we developed a SERS detection platform based on a photo-coupled reaction to enable highly sensitive detection of SA- β -gal in the serum of mice with colitis. First, spherical gold nanoparticles (Au NPs) were prepared using the seed-growth method and sequentially modified with 4-aminophenylthiophenol (PATP) and 4-mercaptobenzenboronic acid (MPBA) on their surfaces to prepare the SERS probe (Scheme 1A). β -FeOOH nanorods were synthesized via the hydrothermal method and reduced to Fe_3O_4 NRs, which were then sequentially assembled with MPBA and β -lactose molecules to produce capture probes with magnetic separation capabilities (Scheme 1B). The β -lactose residue on the surface of the capture probe contained two adjacent cis-hydroxyl structures, which could be selectively recognized by the boric acid group in MPBA, and then drove the SERS probe to be stably bound to the surface of the capture probe through the borate ester bond, thus constructing an SERS detection platform with a core-shell composite structure (Fe_3O_4 NRs@MPBA@ β -lactose-Au NPs@PATP/MPBA). During actual detection, the target SA- β -gal was able to specifically hydrolyze the galactosidic bond in β -lactose, causing the SERS probe to dissociate and be released from the surface of the capture probe. This resulted in a significant reduction in the Raman signal at 1397 cm^{-1} (Scheme 1C). Notably, upon laser irradiation, PATP molecules undergo a photocoupling reaction on the material surface and are converted into 4,4'-dithioazobenzene (DMAB); its N = N vibrational peak (1397 cm^{-1}) is selected as the characteristic reporting signal for quantitative analysis.

2. Experimental

2.1. Materials and reagents

Iron (III) trichloride hexahydrate ($\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$), auric chloride (HAuCl_4), hydrazine hydrate ($\text{N}_2\text{H}_4 \cdot \text{H}_2\text{O}$), tannic acid, p-aminothiophenol (PATP), 3-mercaptopbenzeneboronic acid (MPBA), senescence-associated β -galactosidase (SA- β -gal) was purchased from Aladdin Reagents. Sodium citrate, potassium carbonate (K_2CO_3), and anhydrous ethanol were purchased from Sinopharm Group Pharmaceutical Co., Ltd. β -lactose was purchased from McLean Reagents. The SA- β -gal activity assay kit was purchased from Beijing Box Biotech Co., Ltd. (AKSU042M).

2.2. Instruments and Equipment

The optical properties of the nanomaterials were measured using a UV-visible spectrophotometer (Cary 60). The morphology of the nanomaterials was characterized using a field-emission scanning electron microscope (S-4800 II), a transmission electron microscope (JEM-2100), and a field-emission high-resolution transmission electron microscope (Tecnai F30). Raman signals were acquired using a confocal Raman spectrometer (inVia Qontor). The signal acquisition setup utilized a 50 $\times$ long working distance objective, a 785 nm laser as the excitation source, and a single integration time of 5 s.

2.3. Animal Experiments

This animal experiment protocol has been approved by the Yangzhou University Animal Ethics Committee (202607060). Nine-week-old SPF-grade male C57BL/6 mice were selected, and modeling began after a 7-day acclimatization period. For modeling, a 3% (w/v) sodium dextran sulfate (DSS, molecular weight 36,000–50,000 Da, MP Biomedicals) solution was used in place of regular drinking water, and the mice were allowed to drink ad libitum for 7 days. The DSS solution was freshly prepared daily by dissolving 3 g of DSS powder in 100 mL of sterile distilled water, stirring with a magnetic stirrer until completely dissolved, and then transferring it to light-protected water bottles for the mice to drink. The DSS solution was replaced daily at 9:00 a.m. Tissue samples were collected on Day 7 of the acute phase following the conclusion of DSS induction. During sampling, peripheral blood was first collected; after standing at room temperature for 30 min, it was centrifuged at 3,000 rpm for 15 min to separate the supernatant serum, which was then aliquoted and stored at -80°C for later use. Next, the entire colon was removed. After measuring its length, the distal colon (the segment approximately 0.5 cm from the anal end) was excised. The contents were gently rinsed with pre-chilled PBS buffer and immediately placed in a 4% paraformaldehyde solution for 24-h fixation, in preparation for subsequent paraffin embedding and HE staining analysis. This study's animal procedures follow the AVMA veterinary guidelines: mice were anesthetized with 1% pentobarbital sodium (100 mg/kg) via intraperitoneal injection, and euthanasia was performed using medical-grade CO_2 inhalation.

2.4. Preparation of Fe_3O_4 NRs

According to previous reports, β -FeOOH nanorods were prepared using the hydrothermal method[32]. The specific steps are as follows: Weigh 0.2 g of $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ and measure out 15 mL of ultrapure water; add them sequentially to a 50 mL empty beaker. Place the beaker on a magnetic stirrer and stir continuously for 45 min to obtain a homogeneous aqueous solution of Fe^{3+} with a concentration of 25 mM. The resulting solution was then transferred to a high-pressure reactor and heated at 80°C for 12 h. After the reactor had cooled naturally to room temperature, the yellow solid precipitate at the bottom of the reactor was collected, washed three times with ultrapure water, dried, and stored in a sealed container for later use.

Fe_3O_4 NRs were prepared via a reduction reaction using β -FeOOH as the precursor. The specific procedure is as follows: The yellow β -FeOOH powder prepared as described above was redispersed in 15 mL of ultrapure water; 75 μL of $\text{N}_2\text{H}_4 \cdot \text{H}_2\text{O}$ solution was added to the mixture, which was then placed in an oven set to 80°C for a 4-h reaction. After cooling to room temperature, the yellow product was collected using a magnet, washed three times with ultrapure water, dried, and stored in a sealed container for future use.

2.5. Preparation of SERS Probes

Spherical gold nanoparticles (Au NPs) with a diameter of 5 nm were prepared using the sodium citrate reduction method. Take a 200 mL empty beaker and add the following in sequence: ultrapure water (15.1 mL), sodium citrate (1%, 4 mL), tannic acid (1%, 0.7 mL), and K_2CO_3 (0.1 M, 0.2 mL). Stir continuously to ensure thorough mixing, and set the preheating temperature of the mixed solution to 60°C. Take 1 mL of HAuCl_4 (1%) and add 79 mL of ultrapure water; stir to mix thoroughly to prepare a HAuCl_4 solution with a final concentration of 0.125 mg/mL (dilution ratio 1:80). Set the preheating temperature to 60°C. Quickly add the diluted HAuCl_4 in a single batch to the above mixed solution while stirring continuously. Continue stirring and heating; after boiling for 8 min, the solution will turn a wine-red color. At this point, stop heating and allow the solution to cool naturally to room temperature, yielding Au NPs with a diameter of 5 nm.

10 μL of PATP (1 mM) and 10 μL of MPBA (0.5 mM) were premixed; this premix was then rapidly added to 20 mL of Au NPs solution, which was vigorously stirred for 40 min to promote the formation of Au-S bonds. After the reaction was complete, the solution was washed three times with sodium citrate as the solvent to remove unbound small molecules. Following centrifugation (3000 r/min, 5 min), the sample was resuspended in ultrapure water for later use, yielding the SERS probe (Au NPs@PATP/MPBA).

2.6. Construction of SERS detection platform

Equal volumes of Fe_3O_4 NRS (10 mL) and MPBA (1 mm, 10mL) were left at room temperature for 24 h to complete the grafting of boric acid groups on the surface, and then washed three times with anhydrous ethanol as solvent to remove the physically adsorbed small molecules. Then, the above products were transferred to β -lactose (50 mm, 10mL) and incubated for 3h to fully combine the boric acid group with sugar molecules to prepare the capture probe (Fe_3O_4 NRs@MPBA@ β -lactose).

Secondly, the SERS probe and the capture probe were mixed in equal volume, reacted for 40 min, and then centrifuged and purified in the same way to obtain the composite structure (Fe_3O_4 NRs@Au NPs), SERS detection platform was successfully constructed.

2.7. Raman spectrum acquisition

The sample to be tested (8 μL) was mixed with SERS detection platform (1 mL) and incubated for 35 min. After the reaction, the products were enriched and separated by micro magnet, and washed several times with PBS buffer for SERS detection. The spectral data were smoothed and baseline processed, and the average of three independent measurement results was taken for each curve.

3. Results and discussions

3.1. Characterization of probes

The capture probes and SERS probes were characterized using electron optical techniques such as scanning electron microscopy (SEM) and transmission electron microscopy (TEM). As shown in Fig. 1A and Fig. 1B, Fe_3O_4 NRs exhibited a distinct rod-like structure with an average length of approximately 165 nm and a width of approximately 45 nm. As shown in Fig. 1C, the Au NPs exhibited a distinct spherical structure in the past, with uniform sizes and an average diameter of approximately 5 nm. The UV-visible absorption spectra of the Au NPs and Fe_3O_4 NRs were recorded (Fig. 1D); the UV absorption peaks of the Au NPs and Fe_3O_4 NRs were located at 532 nm and 518 nm, respectively.

3.2. Characterization and Performance Evaluation of a SERS Detection Platform

This study conducted a multidimensional validation of the morphological characterization, elemental distribution, optical properties, and electric field enhancement effects of the constructed SERS detection platform (Fe_3O_4 NRs@MPBA@ β -lactose-Au NPs@PATP/MPBA). As shown in Fig. 2A and Fig. 2B, Au NPs were densely assembled on the surface of Fe_3O_4 NRs. The SERS detection platform showed an obvious core-shell composite structure, with an average length of 175 nm and a width of about 55nm. The element mapping of the SERS detection platform was characterized. As shown in Fig. 2C and Fig. 2D, the SERS detection platform took Fe_3O_4 NRS as the core and Au NPs as the shell. As shown in Fig. 2G, after co-incubation of the capture probe (Fe_3O_4 NRs@MPBA@ β -lactose) with the SERS probe (Au NPs@PATP/MPBA), a distinct Raman peak at 1070 cm^{-1} due to MPBA modified onto the surface of Fe_3O_4 NRs via Au-S bonding (black curve). The β -lactose molecule contained CIS diol structure, which could specifically bind to the boric acid group of MPBA. This combination did not result in any significant change in the SERS signal of Fe_3O_4 NRs@MPBA@ β -lactose (red curve). However, when the capture probe was further incubated with the SERS probe (blue curve), the SERS probe would specifically bind to β -lactose and then assemble on the surface of Fe_3O_4 NRS to form Fe_3O_4 NRs@MPBA@ β -lactose-Au

NPs@PATP/MPBA composite structure as SERS detection platform. Notably, three newly emerging characteristic peaks at 1140 cm^{-1} , 1397 cm^{-1} , and 1456 cm^{-1} in the Raman spectra of the Fe_3O_4 NRs@MPBA@ β -lactose-Au NPs@PATP/MPBA composite structure were attributed to the catalytic reaction of PATP molecules on the material surface and their subsequent conversion to DMAB. Accordingly, this study selected the N = N vibrational signal in DMAB as the detection indicator[33]; the corresponding Raman peak was located at 1397 cm^{-1} . In this study, the LSPR enhancement effects and signal amplification capabilities of the composite structure of the SERS detection platform were evaluated through electromagnetic field simulations and spectroscopic experiments. As shown in Fig. 2H, by analyzing the Raman spectra of pure DTNB (10^{-1} M) and the DTNB-labeled SERS detection platform (10^{-6} M), using the Raman characteristic peak at 1342 cm^{-1} as a reference, the enhancement factor (EF) was calculated to be 1.29×10^6 . Simulations using the finite-difference time-domain (FDTD) method revealed that, in the XY cross-section (Fig. 2I and Fig. 2K), when incident light struck along the plane of the long axis and was p-polarized along the X direction, the maximum electric field intensity E_{max} was 3.87, and the electric field enhancement was primarily concentrated in the gaps between the gold nanoparticles surrounding the rod-like structure. In the XZ cross-section (Fig. 2J and Fig. 2L), when the incident light was incident perpendicularly and the p-polarization was perpendicular to the cross-sectional direction, the maximum electric field intensity E_{max} was 2.7, and the electric field enhancement was concentrated in the gaps between the nanoparticles at both ends of the rod-like structure. The above results indicate that the SERS detection platform exhibits excellent SERS activity.

3.3. Condition optimization

This study optimized parameters such as β -lactose concentration, pH, and target incubation time, significantly improving the analytical reliability and stability of the SERS detection platform. As shown in Fig. 3A, as the concentration of β -lactose increased, the number of SERS probes bound to the surface of the capture probes also increased. As the assembly density of Fe_3O_4 NRs@MPBA@ β -lactose-Au NPs@PATP/MPBA increased, the number of formed Raman “hotspots” also increased, leading to a significant upward trend in SERS signal intensity. When the β -lactose concentration reached 40 mM, the binding sites on the surface of the capture probe approached saturation. At this point, the signal intensity of the SERS characteristic peak no longer continued to increase with rising concentration and instead leveled off. Since the DMAB formation process during the PATP coupling reaction is a catalytic reaction dependent on the acid-base environment, this study further optimized the pH conditions. As shown in Fig. 3B, as the pH gradually increased from 6.0 to 7.5, the Raman signal intensity increased significantly with rising pH. This is attributed to the fact that a weakly alkaline environment promotes the catalytic dehydrogenation and coupling of PATP on the surface of Au NPs, thereby generating more DMAB molecules with strong N = N vibrational signals. When the pH reached 7.5, the signal intensity peaked. Further increasing the pH to 8.0 did not result in any significant change in the signal, indicating that the catalytic conversion had reached saturation. Therefore, a pH of 7.5 was selected as the optimal reaction condition for the SERS detection platform. As shown in Fig. 3C, as the target incubation time

increased, the intensity of the characteristic Raman peak at 1397 cm^{-1} exhibited a gradual decline, and the signal intensity stabilized after 50 min. This change resulted from the complete degradation of β -lactose under the catalytic action of SA- β -gal, which disrupted the original $\text{Fe}_3\text{O}_4\text{ NRs@MPBA@}\beta\text{-lactose-Au NPs@PATP/MPBA}$ composite structure and caused the SERS probe to gradually detach from the sensing interface, thereby reducing the LSPR enhancement to its minimum level. Therefore, in this study, the β -lactose concentration was ultimately set to 40 mM, and the system pH was adjusted to 7.5. After 50 minutes of incubation with the target, these conditions were established as the optimal experimental conditions and were used for the subsequent performance evaluation of the SERS detection platform.

3.4. Performance evaluation

Based on the optimal experimental parameters described above, the analytical performance of the SERS detection platform was further evaluated, specifically including specificity, reproducibility, and detection sensitivity. Specificity and reproducibility are key factors affecting the reliability and stability of detection results. In this study, AMY, IgG, HAS, and blank were selected as the control groups, and SA- β -gal (10 U/mL) was used as the target group. The corresponding SERS detection results were shown in Fig. 4A and Fig. 4B. Using the Raman characteristic peak at 1397 cm^{-1} corresponding to the N = N vibrational mode in DMAB, the intra-assay relative standard deviation (RSD) for the control group was calculated to be 6.57%, indicating that the SERS detection platform exhibited excellent target-specific recognition selectivity for SA- β -gal and could effectively avoid the influence of other interferents. To verify the reproducibility of the platform's results, five independently prepared batches of the SERS detection platform were selected and used to perform parallel assays on a SA- β -gal standard solution of the same concentration (10 U/mL) under identical experimental conditions. The corresponding SERS detection results were shown in Fig. 4C and Fig. 4D. Calculations showed that the RSD value among the five batches of SERS detection platforms was 8.32%, indicating that the SERS detection platform possessed satisfactory inter-batch consistency and detection stability.

3.5. Quantitative assay of SA- β -gal

Sensitivity is directly related to the quantitative analysis capability and trace detection limit of SERS detection platforms for SA- β -gal. SA- β -gal standards were dissolved in fetal bovine serum matrix to prepare concentration gradient solutions covering a range from 10^{-4} to 10 U/mL. As shown in Fig. 5A, as the concentration of the target SA- β -gal increased, more SA- β -gal specifically hydrolyzed the β -galactose in the SERS detection platform, causing the β -galactoside bonds in the $\text{Fe}_3\text{O}_4\text{ NRs@MPBA@}\beta\text{-lactose-Au NPs@PATP/MPBA}$ composite structure to break. The SERS probes then detached from the surface of the capture probes, resulting in a decrease in the signal intensity of the Raman characteristic peak at 1397 cm^{-1} . As shown in Fig. 5B, when the logarithm of the SA- β -gal concentration was plotted on the x-axis and the response value of the characteristic peak at 1339 cm^{-1} was plotted on the y-axis, the fitted curve exhibited a good linear fit. The fitted calibration curve is $y = 1436.23x + 8199.12$ ($R^2 = 0.970$), and

calculations show that the detection limit (LOD) of this platform is 1.956×10^{-6} U/mL. A comparison of the analytical performance of the SERS detection platform with existing methods (Table 1) shows that this platform not only has significantly higher sensitivity than the comparison methods but also has a substantially broader detection range, demonstrating excellent broad-concentration response capability.

Table 1
Comparison of the SERS detection platform with other detection methods

Method	Biomarker	Linear range	Detection limit	Reference
Colorimetric	β-gal	5×10^{-5} - 5×10^{-3} U/mL	4×10^{-5} U/mL	[34]
Fluorescence	β-gal	2×10^{-4} - 5×10^{-2} U/mL	3.1×10^{-5} U/mL	[35]
Immunoassay	β-gal	0 - 0.22 U/mL	1.69×10^{-3} U/mL	[36]
SERS	SA-β-gal	10^{-4} - 10 U/mL	1.956×10^{-6} U/mL	This work

3.6. Clinical sample characterization

To validate the efficacy of the DSS-induced colitis model used in this study, we compared the gross colonic morphology, histopathological changes (Fig. 6A), and body weight trends (Fig. 6B) among mice in the control group and those in the mild and severe colitis groups. Macroscopic examination revealed that the control group had a normal colon length and a smooth intestinal wall; the mild group exhibited a slightly shortened colon and a thickened intestinal wall with mild congestion; the severe group showed a significantly shortened colon, severe congestion and edema of the intestinal wall, and intestinal malformation, presenting inflammation-dependent changes. HE staining results further indicated that the control group had an intact mucosa with regular crypt structures; in the mild group, the mucosa showed mild erosion and focal inflammatory infiltration; and in the severe group, the mucosa exhibited extensive erosion, massive destruction of crypts, and diffuse infiltration of inflammatory cells, consistent with the typical pathological features of severe colitis. Body weight monitoring showed that in the control group, weight increased steadily throughout the study; in the mild group, weight began to decline slowly after Day 3 and stabilized at 18.5–19.0 g by Day 7; in the severe group, body weight declined rapidly and continuously after Day 2, reaching its lowest point (approximately 16.42 g) on Day 6, which closely correlated with the severity of inflammation. These results indicate that DSS treatment successfully established a colitis model with clear gradient differences (Severe > Mild > Control), providing a reliable animal model foundation for subsequent in vivo evaluation of the SERS platform’s detection performance.

3.7. Testing of actual serum samples

Using a SERS detection platform, serum samples from healthy mice, mice with mild colitis, and mice with severe colitis were analyzed to obtain the average SERS spectra of mouse serum under these three

different disease states (Fig. 7A). As shown in Fig. 7B, the average SERS signal intensity in mouse serum exhibited a gradual decrease as the severity of colitis increased. By substituting the intensities of the characteristic peak at 1397 cm^{-1} from the average SERS spectra of serum samples from healthy mice, mice with mild colitis, and mice with severe colitis into the linear regression equation (Fig. 5B), we calculated the SA- β -gal expression levels in mouse serum for the three disease states (Table S1). We then compared these results with those obtained using the micro-colorimetric assay (Table 2). the experimental results showed a high degree of consistency, with relative errors all less than 10%.

Table 2
Comparison of SA- β -gal expression levels in mouse serum from different disease states using the SERS detection platform and the colorimetric method.

Sample	SERS (U/mL)	Colorimetric (U/mL)	Relative error (%)
Control	3.02	2.90	-3.98
Mild colitis	18.02	17.00	-5.67
Severe colitis	40.34	42.02	4.18

4. Conclusion

In summary, this study successfully established a SERS detection platform based on a magnetic core-shell composite structure (Fe₃O₄ NRs@MPBA@ β -lactose-Au NPs@PATP/MPBA) to detect SA- β -gal expression levels in serum with high sensitivity, thereby enabling the early diagnosis of colitis. This study leveraged the magnetic separation and enrichment capabilities of Fe₃O₄ NRs and the localized surface plasmon resonance enhancement properties of Au NPs, and, based on the borate-specific recognition between MPBA and β -lactose, successfully established a SERS detection platform based on a photo-coupled reaction. This detection platform not only combines excellent magnetic separation efficiency with signal enhancement capabilities but also demonstrates good specificity and reproducibility in complex serum matrices, effectively reflecting changes in SA- β -gal levels in mice with a colorectal inflammation model. Compared with traditional detection methods, this platform exhibits significant advantages in both detection limit and dynamic response range, providing reliable technical support for the point-of-care detection of biomarkers associated with inflammatory bowel disease.

Declarations

Declaration of Competing Interest

The authors declare that there are no conflicts of interest related to this article.

Data availability

Data can be provided as required.

Disclosure of AI tool usage

No generative AI tools were used to generate the core experimental data, findings, or original academic views in the preparation of this manuscript.

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Author Contribution

Xinmei Zheng and Min Xu wrote the main text of the manuscript; Yanwen Zhuang, Yangfan Zhou, and Yijiang Wu completed the data visualization and methodology; Hui Chen provided supervision and project management; and Yan Wang provided funding.

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Schemes

Scheme 1 is available in the Supplementary Files section

Figures

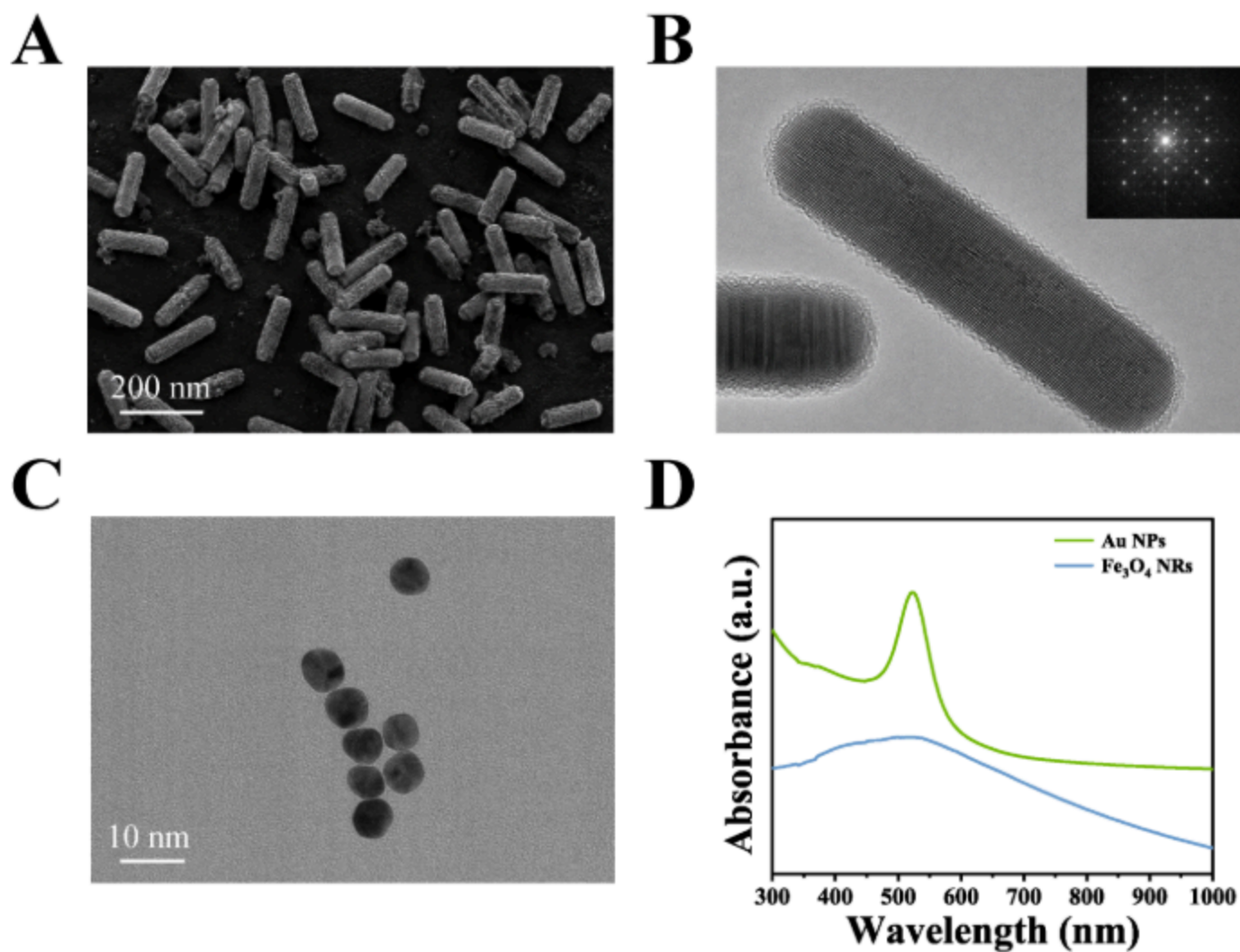


Figure 1

(A) SEM image of Fe₃O₄ NRs; (B) TEM and SAED images of Fe₃O₄ NRs; (C) TEM image of Au NPs; (D) UV-visible absorption spectra of Au NPs and Fe₃O₄ NRs.

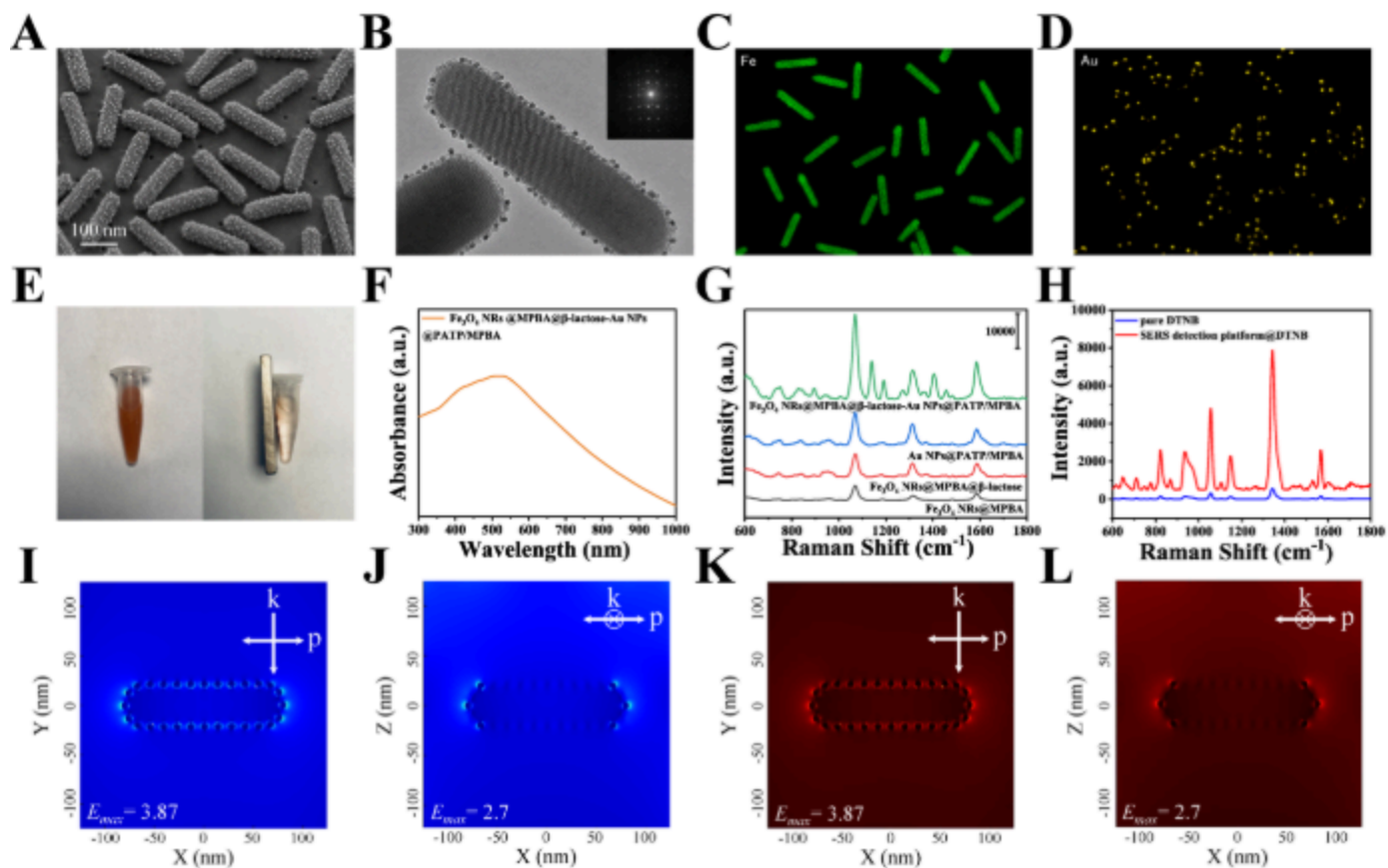


Figure 2

(A) SEM image, (B) TEM image and SAED image, (C–D) elemental mapping images, (E) image of the magnetic aggregation process, (F) UV-visible absorption spectrum, (G) spectral changes during the preparation process, (H) evaluation of signal enhancement capability, and (I and K) simulated electric field distribution in the X-Y plane for the SERS detection platform. (J and L) Simulated electric field distribution in the X-Z plane.

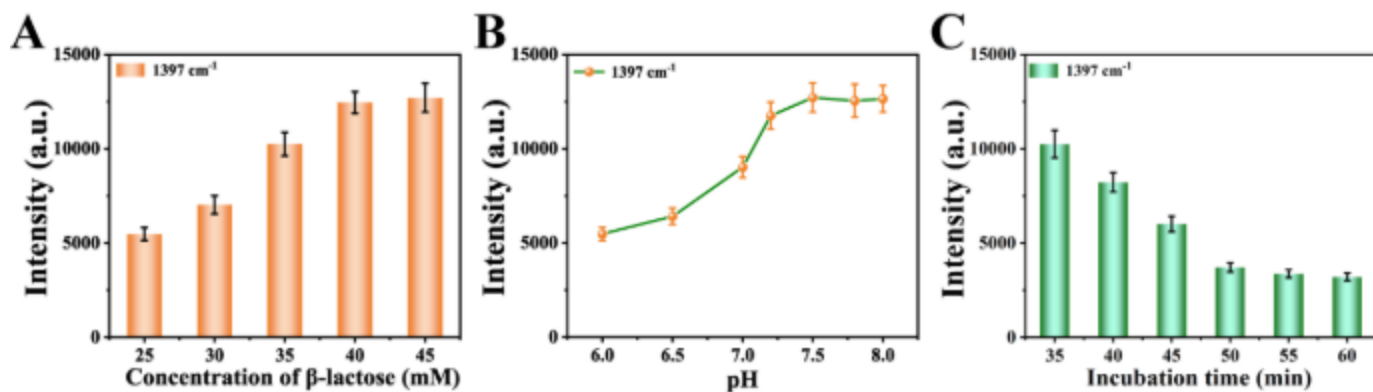


Figure 3

Parameter optimization. (A) β -lactose concentration, (B) pH, and (C) incubation time.

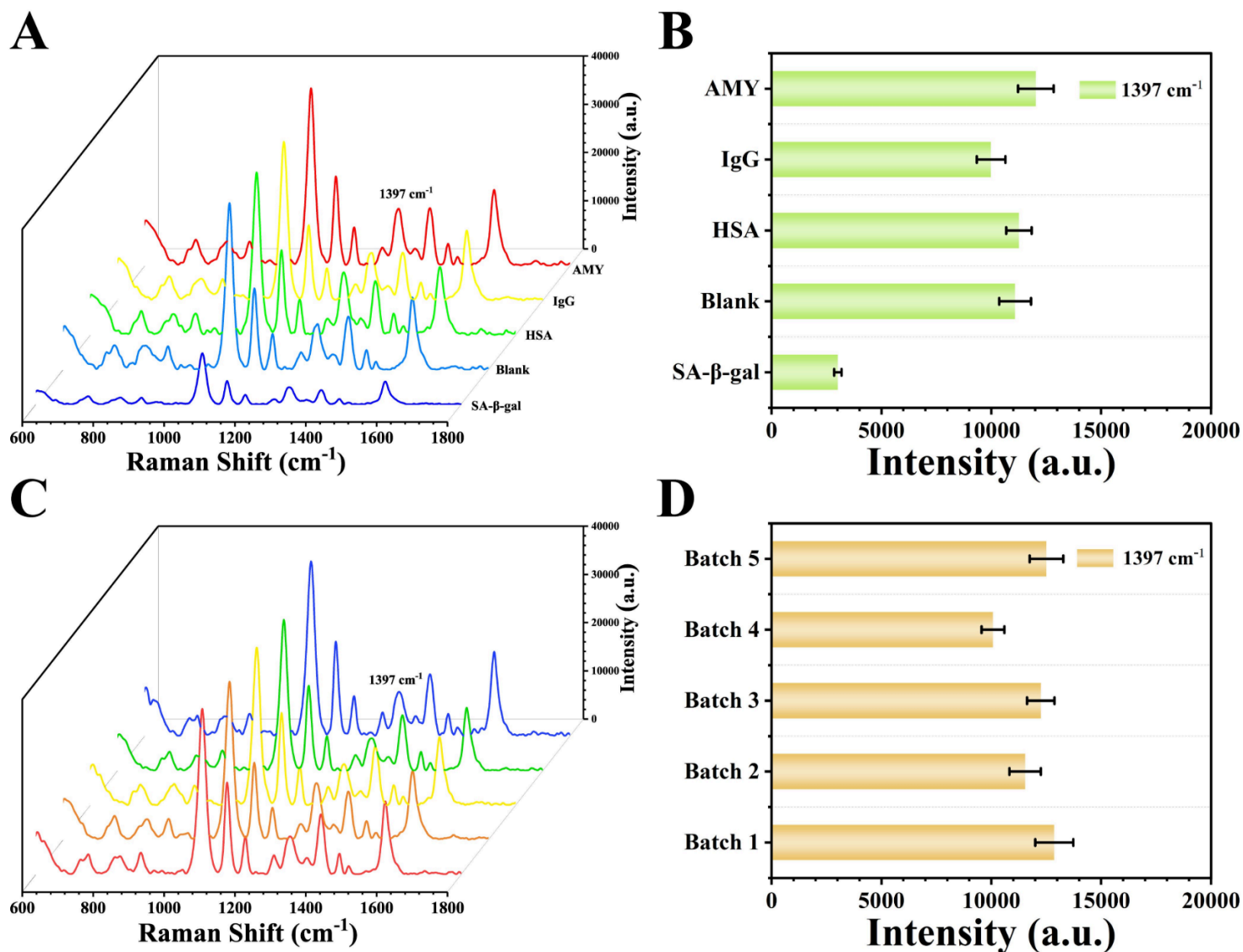


Figure 4

(A) Evaluation of the specificity of the SERS detection platform and (B) a bar chart showing the signal intensity of the characteristic Raman peak at 1397 cm^{-1} . (C) Evaluation of the reproducibility of the SERS detection platform and (D) a bar chart showing the signal intensity of the characteristic Raman peak at 1397 cm^{-1} .

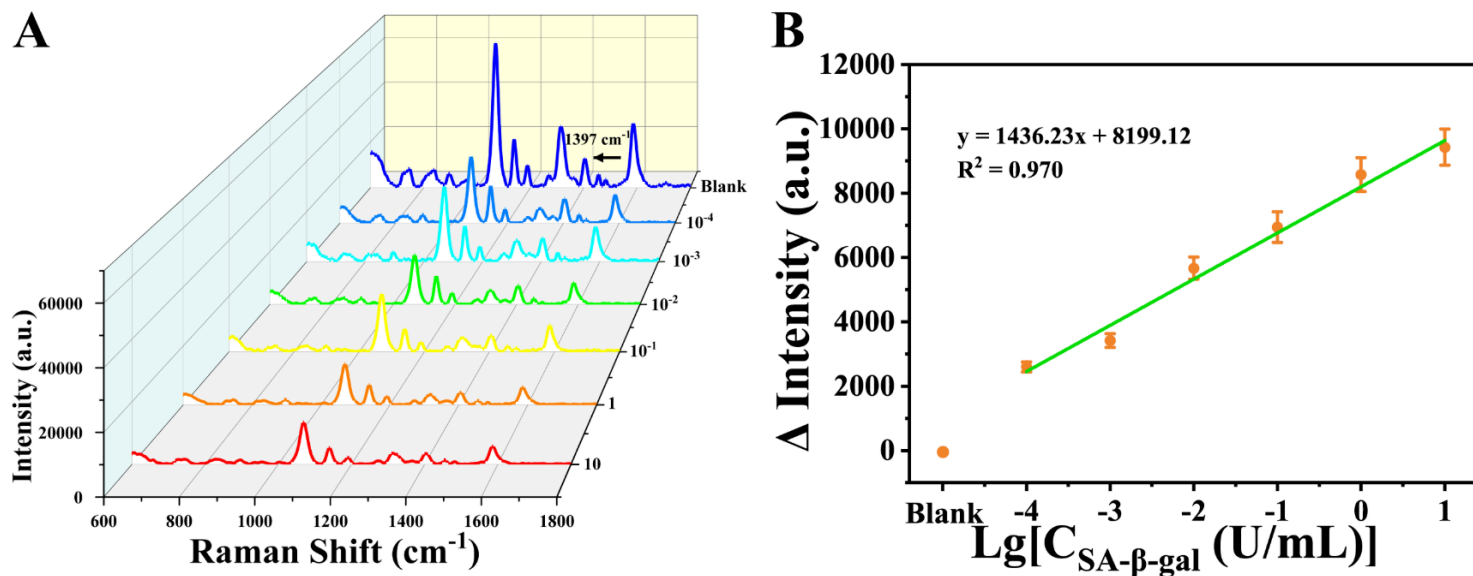


Figure 5

(A) Detection of the Raman spectra of SA- β -gal at different concentration gradients in serum using the SERS detection platform. (B) Calibration curve showing the relationship between the signal intensity of the characteristic Raman peak at 1397 cm^{-1} and the logarithm of the SA- β -gal concentration.

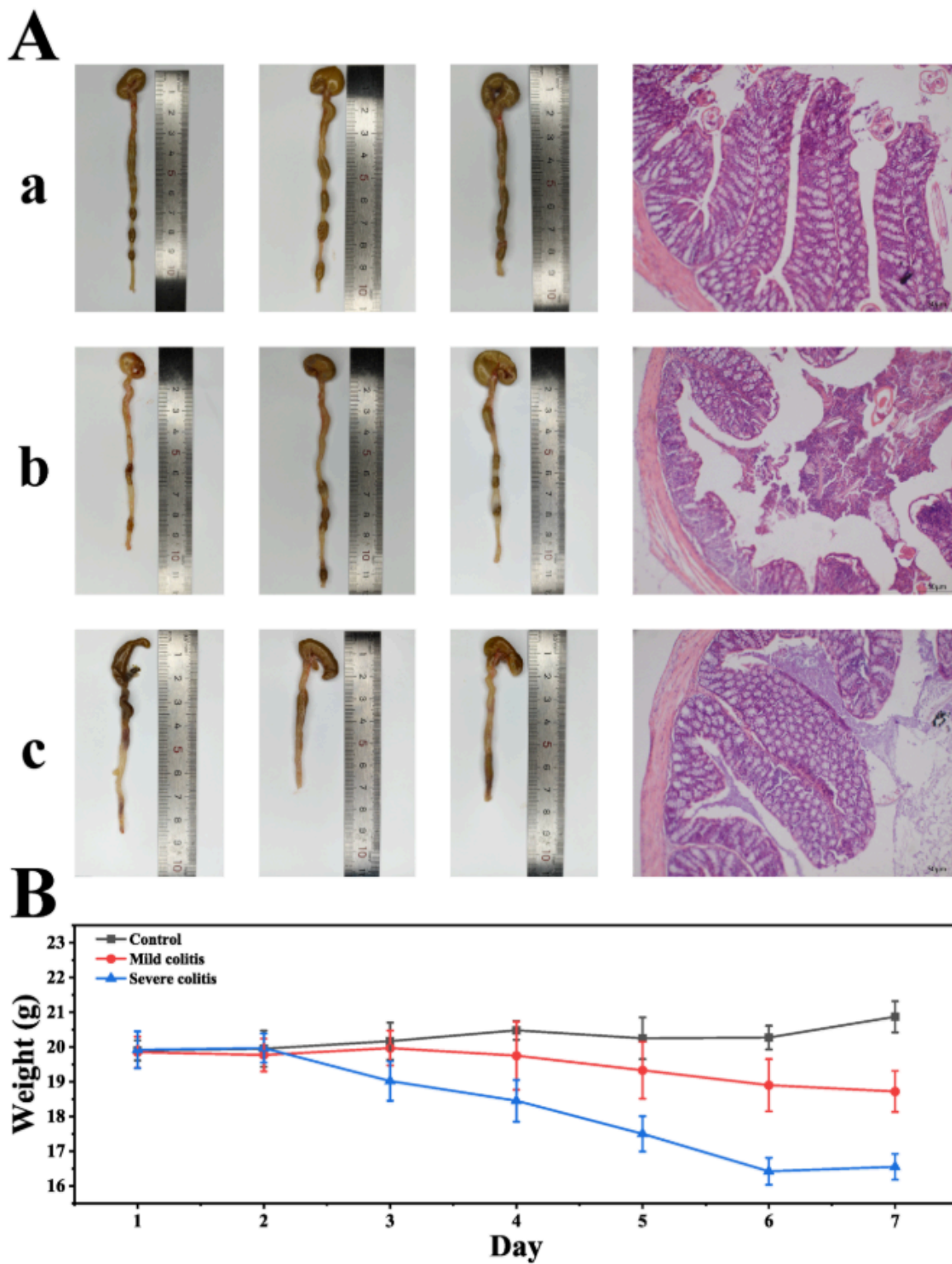


Figure 6

(A) Gross specimens of the colon and histopathological sections of intestinal tissue: a: Control; b: Mild colitis; c: Severe colitis. (B) Curves showing changes in body weight over time in mice.

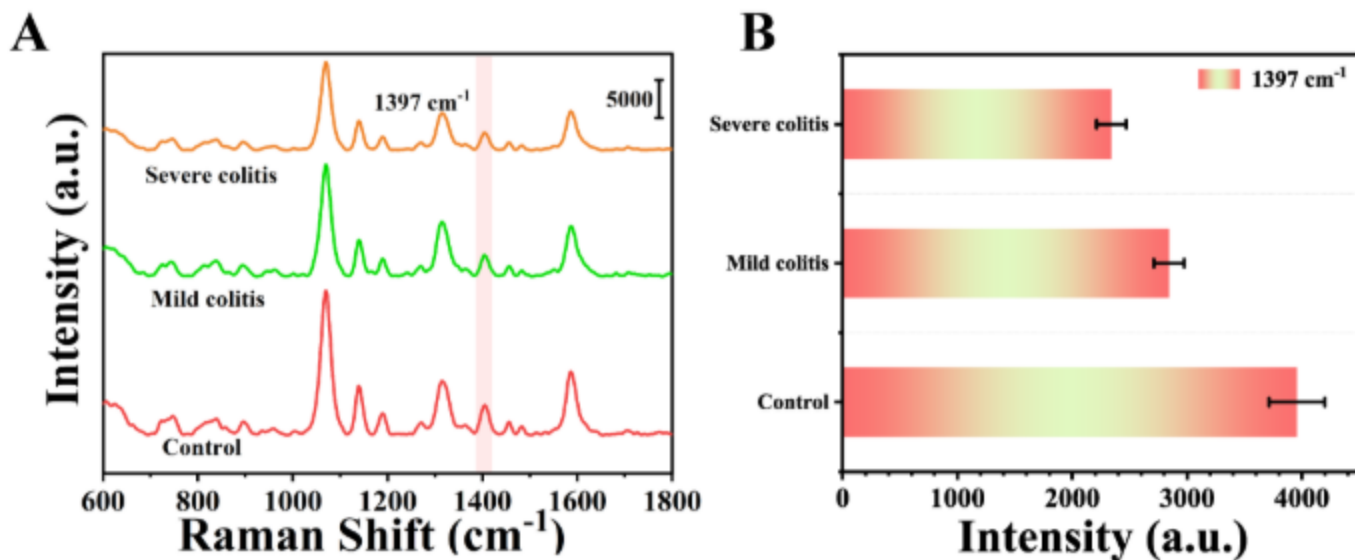


Figure 7

(A) Average SERS spectra of serum from healthy mice, mice with mild colorectal inflammation, and mice with severe colorectal inflammation. (B) Bar chart showing SERS intensities corresponding to the Raman feature peak at 1397 cm⁻¹.

Supplementary Files

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