Research article

Raman enhancement effect of different silver nanoparticles on salbutamol

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HIGHLIGHTS

- Raman enhancement signal of salbutamol was compared with concentrated gold and silver colloids.
- The effect of silver nanoparticles sizes on the enhancement effect are in particular broached.
- The methods can realize salbutamol at trace concentrations detection.

ARTICLE INFO

ABSTRACT

Salbutamol is a β-adrenergic receptor agonist compound which has been abused as an animal growth promoter to improve carcass lean meat percentage. At present, the detection of salbutamol by SERS mostly uses gold colloid as substrate, which is expensive and has a high detection limit. In this report, Raman enhancement signal of salbutamol was compared with concentrated gold and silver colloids. The results show that the concentrated silver colloid prepared by reducing silver nitrate with hydroxylamine hydrochloride had superior performance. Three silver colloids with different particle sizes were synthesized by the same reducing agent and used as substrates for spectra acquisition of salbutamol to explore the enhancement performance of different silver nanoparticles sizes on salbutamol. The results showed that silver nanoparticles with larger particle sizes were more conducive to the adsorption of salbutamol. Finally, under the optimal conditions (Silver colloid A as enhanced substrate, 0.2 mol/L NaOH aqueous solution as aggregating compound), a better linear relationship between the concentration of salbutamol (ranged from 0.2 to 1 mg/L) and SERS intensity. The linear equation between SERS intensity and salbutamol concentration was $C = 0.0023 \bullet I-0.079$ (mg/L) with a good linearity ($R^2 = 0.994$) and lower root mean square error ($RMSE_c = 0.022$ mg/L), where C (mg/L) was the concentration of salbutamol solution and I was the SERS intensity of salbutamol solution. Validation set correlation coefficient was 0.988 and prediction root mean square error was 0.029 mg/L. This method provides a new idea for further reducing the detection limit of salbutamol. This study is helpful to further develop a simple and low-cost SERS detection method of salbutamol based on silver colloid.

1. Introduction

Salbutamol (SAL) is a phenol-β_2-agonist which is widely used for the treatment of human respiratory diseases (Dunshea, 1993; He et al., 2020a). In addition, it also acts as a nutrient repartitioning agent and has been used to produce leaner meat with a higher muscle to fat ratio for livestock (Boler et al., 2012; Cheng et al., 2016; Mohamed et al., 2020). If livestock and poultry are fed a large amount of salbutamol, most of it will be deposited in the muscle and liver of animals for a long time (Chan et al., 2016). Food contaminated with salbutamol can cause heart palpitations, muscle pain, dizziness, vomiting, kidney damage and even life

threatening (Zhang et al., 2007). Laws were established in The European Union, the United States, Japan and other countries to restrict the use of stimulants promoting animal growth as feed additives in livestock and poultry production, and all stipulate that salbutamol should not be detected (Stolker and Brinkman, 2005).

There are various methods to detect and control the illegal sale of fresh meat containing salbutamol. So far, the traditional detection methods are based in high performance liquid chromatography (HPLC) (Morales-Trejo et al., 2013; Yan et al., 2016), liquid chromatography-mass spectrometry (LC-MS) (Chang et al., 2018), gas chromatography-mass spectrometry (GC-MS) (Jouyban et al., 2020), and

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Enzyme-linked immunosorbent assay (ELISA) (He et al., 2020b; Zhao et al., 2020), etc. However, HPLC, LC-MS and GC-MC all require expensive equipment, professional operators, complex operating procedures and a lot of time. On the other hand, Enzyme-linked immunosorbent assay (ELISA) requires the preparation of expensive antibodies and is prone to inactivation, resulting in unstable results.

Surface-enhanced Raman spectroscopy (SERS) is a novel analytical technique for trace contaminant determination which was realized by the enhanced local electromagnetic field near the nanostructured noble metal surfaces (Bi et al., 2022; Panneerselvam et al., 2018). SERS technology can enhance the Raman signal intensity of target molecules by 10^4–10^6 times (Lam et al., 2020). There were some reports to detect β_2-agonist by SERS. Zhai et al. detected ractopamine in swine urine with a detection limit of 0.8 mg/L using SERS (Zhai et al., 2011). Lorenzo et al. investigated the absorption of salbutamol on Au and Ag nanoparticles at different pH values and employed SERS to detect salbutamol. The results showed that the enhancement signal of salbutamol with gold colloid was better than silver colloid as substrate, and the detection limit of salbutamol was 0.765 mg/L (Izquierdo-Lorenzo et al., 2010). The enhancement of β_2-agonist by gold colloid as enhancement substrate has been reported in the above studies, but there are few reports about silver colloid. There are many factors affecting the adsorption of metal nanoparticles on target molecules, such as the shape, size of metal nanoparticles and the use of aggregating compounds, and so on (He et al., 2017). Ngoc Pham et al. carried out the synthesis of multi-shaped silver nanoparticles from the silver nanospheres seeds via the etching-growth process for the detection of salbutamol. The findings demonstrated that multi-shaped silver nanoparticles had a better enhancement effect on salbutamol with a detection limit of 1.25 mg/L (Pham et al., 2020). Better enhancements for salbutamol need to be further investigated.

In this report, concentrated silver colloid was used as substrate for the detection of salbutamol which has a better SERS effect; In addition, different types of colloids were used for detection of salbutamol and it was found that the silver colloid prepared by reducing silver nitrate with hydroxylamine hydrochloride had better enhancement performance on salbutamol; Then, the enhancement effect of several silver colloids with different particle sizes as substrates for detection of salbutamol was compared; finally, the influence of different concentrations of sodium hydroxide solution as aggregating compounds for SERS effect were compared, and the optimal detection conditions were established.

2. Materials and methods

2.1. Materials

Chloroauric acid (HAuCl$_4$•4H$_2$O) and silver nitrate (AgNO$_3$) were purchased from Shenyang Jinke Reagent Co., LTD and Sinopharm Group Chemical Co. LTD respectively. Salbutamol was purchased from the National Institute for Food and Drug Control. Hydroxylamine hydrochloride, sodium citrate and sodium hydroxide were obtained from Shanghai Maclean Technology Co., LTD. (Shanghai, China), and all the chemicals were analytical grade. All reagents dissolved and diluted with ultrapure water.

2.2. Synthesis of silver and gold colloids

Five different kinds of silver colloids and three different sizes of gold colloids were synthesized in this report, herein referred to as silver colloids A~E and gold colloids F ~ H. Using hydroxylamine hydrochloride and sodium hydroxide mixed solution as reducing agent, silver colloids A, B and C were prepared by changing the mixing sequence and mixing rate of silver nitrate and hydroxylamine hydrochloride/sodium hydroxide solution in silver colloid synthesis. Silver colloids D and E were prepared by reducing silver nitrate with sodium citrate and sodium borohydride, respectively. Gold colloids F, G and H with different particle sizes were prepared by adding different amounts of sodium citrate to

reduce chloroauric acid. All the prepared gold and silver colloids were centrifuged and concentrated at 3000 r/min in 20 min. The supernatant was removed, and the precipitated nanoparticles were placed in a dark environment at 4 °C for future use.

2.2.1. Synthesis of silver colloids

Silver colloid A was prepared by dropping the mixed solution of hydroxylamine hydrochloride and sodium hydroxide into silver nitrate solution (Leopold and Lendl, 2003). Briefly, A silver nitrate solution was prepared by adding 0.017 g AgNO$_3$ to 90 ml ultrapure water (1.11 × 10^{-3} M AgNO$_3$ solution, solution I). A mixture of hydroxylamine hydrochloride and sodium hydroxide was prepared by adding 0.01 g hydroxylamine hydrochloride and 0.012 g NaOH to 10 ml ultrapure water (solution II). 10 ml of solution I was added dropwise to 90 mL solution II with intense stirring for 20 min.

Silver colloid B was prepared by adding rapidly the mixed solution of hydroxylamine hydrochloride and sodium hydroxide into silver nitrate solution (Leopold and Lendl, 2003). Specifically, using solution 1 and solution 2 prepared above, 10ml solution I was added rapidly to 90ml solution II with stirred vigorously for 20 min.

Silver colloid C was prepared by adding rapidly the silver nitrate solution into mixed solution of hydroxylamine hydrochloride and sodium hydroxide (Leopold and Lendl, 2003). Briefly, A silver nitrate solution was prepared by adding 0.017 g AgNO$_3$ to 10 ml ultrapure water (0.01 M AgNO$_3$ solution, solution III). A mixture of hydroxylamine hydrochloride and sodium hydroxide was prepared by adding 0.01 g hydroxylamine hydrochloride and 0.012 g NaOH to 90 ml ultrapure water (solution lV). Afterward, 10 ml of solution III was added rapidly to 90 mL solution lV with intense stirring for 20 min.

The silver colloid D was prepared according to Lee-Meisel method (Lee and Meisel, 1982). A total of 500 mL of 10^{-3} M AgNO$_3$ aqueous solution was carried to boiling. Then, 10 mL of 1% (by weight) trisodium citrate solution was rapidly added to the reaction solution with continuous mechanical stirring. The solution was kept on boiling for circa 1 h and then cooled to a room temperature of 25 °C.

The silver colloid E was produced using sodium borohydride as reducing agent as follows (Creighton et al., 1979): A solution of 10^{-3} M AgNO$_3$ prepared by adding 0.017 g AgNO$_3$ to 100 ml ultrapure water, was added rapidly to 300 mL of vigorously stirred ice-cold 2×10^{-3} M NaBH$_4$ solution. The solutions of these salts were mixed rapidly with vigorous shaking to aid monodispersed and the solutions turned yellow.

2.2.2. Synthesis of gold colloids

Gold colloids F ~ H were synthesized by the reduction of gold chloride with the different volumes of sodium citrate in aqueous solution which brought widely different particle diameters (Frens & Z., 1972). The procedure was described as follows: solutions were prepared of 0.01% gold chloride by weight and of 1% trisodium citrate solution by weight respectively. 500 ml of gold chloride solution was heated to boiling and three different volumes (3 ml, 5 ml and 7.5 ml) of trisodium citrate solution were added respectively. The mixture solution was boiled for approximately 10 min and the boiling solution turns wine red. Finally, three gold nanoparticle solutions with different particle sizes were obtained which were referred to gold colloids F ~ H respectively.

2.3. Spectral collection

Salbutamol solutions with concentrations ranging from 0.1 mg/L to 10 mg/L were prepared by dissolving salbutamol into ultrapure water. NaOH solutions of 0.1, 0.2 and 0.3 mol/L were prepared as aggregating compounds. 2.5 μL silver colloid or gold colloid, 2.5 μL salbutamol aqueous solution and 2.5 μL NaOH solution were dropped sequentially on an aluminum substrate and stood for 10 s before spectra were collected. Each concentration sample was collected three times, and three Raman spectra were collected each time. The spectra obtained were averaged as the original spectra of the sample. UV/vis absorption spectra were

collected by adding 1 mL gold or silver nanoparticles solution (before centrifugation) and 3 mL ultra-pure water in the colorimetric dish.

Raman spectra were acquired by a Raman spectrometer ATP5020 manufactured by Aopu Tiancheng Technology Co., LTD. The Raman excitation source was a laser with a wavelength of 785 nm. Laser power was measured to be 400 mW, and integral time was 5 s. UV/vis absorption spectra were collected by a UV/vis Spectrophotometer 756S, in the 1100–190 nm spectral range. Transmission electron microscopy (TEM) (JEM-1200EX, JEOL Ltd., Tokyo, Japan) was employed to obtain the TEM images of silver colloids.

2.4. Data processing

The pre-processing method of Automatic Whitacre Fitting Algorithm (AWF) was used to remove the fluorescence background of Raman spectra after acquisition. Correlation coefficient (R_c), root mean square error of correction ($RMSE_c$), prediction coefficient (R_p) and root mean square error of prediction ($RMSE_p$) were used to evaluate the performance of the models (Chellini et al., 2017). Higher R_p, R_c values and lower $RMSE_c$, $RMSE_p$ values demonstrate better model performance (Guo et al., 2021). All calculations were performed in (MathWorks. Inc, Natick, America).

3. Results and discussion

3.1. Characterization of the silver and gold colloids

As shown in Figure 1, the optical properties of all the colloidal solutions were characterized by UV/vis absorption spectra. The particle size and dispersity of colloids can be estimated with peak position and peak width (He et al., 2017). The increase of particle size and polydispersity of colloids correspond to the phenomenon of redshift and broadening of UV/vis absorption spectra (Haiss et al., 2007). The UV/vis absorption spectra curves of five kinds of silver colloids were shown in Figure 1. Curves a, b and c were the UV/vis absorption spectra of silver colloids prepared by reducing silver nitrate with hydroxylamine hydrochloride. Curves d and e were the UV/vis absorption spectra of silver colloid prepared by reducing silver nitrate with sodium citrate and sodium borohydride, respectively. The silver colloids B, C, D and E were mono-dispersive which the particle size is relatively concentrated with the maximum absorption bands of 423nm, 409 nm, 417 nm and 393nm, respectively shown in Figure 1(A). A broadening occurred at the absorption peak of the UV-vis absorption spectrum of silver colloid A which indicates that the silver colloid A has polydispersity and large average particle size. This is because the dropwise addition of the reducing agent probably leads to a seeding effect resulting in a growth of the silver

particles. TEM images of silver colloids A~C were shown in Figure 2 and we observed the average particle size of silver colloid was A > B > C.

Gold colloids F ~ H were produced by the reduction of gold chloride with the different volumes of sodium citrate in aqueous solution which brought widely different particle diameters. UV-vis absorption spectra curves of three gold colloid with different particle sizes were shown in Figure 1(B), which were similar to the gold colloid reported in previous studies. From Figure 1(B), we observed the maximum absorption bands of the UV-vis absorption spectra of gold colloids F, G and H were 526nm, 532nm and 540 nm respectively, which indicates that gold colloids F ~ H had the diffident particle diameters.

3.2. Enhanced spectra of salbutamol with different substrates

Due to the small concentration of silver colloid D prepared by sodium borohydride reduction of silver nitrate, no concentrated silver colloid was obtained after centrifugation. Only silver colloids A~D were used as substrates for the test. In order to explore the adsorption effect of different substrates on salbutamol, the SERS spectra of salbutamol solution (10 mg/L) were collected with silver colloids A, B, C, D and gold colloids F, G, H as substrate respectively and 0.2 mol/L NaOH solution as the aggregating compound. At the same time, the same substrate and aggregating agent were used to collect SERS spectra of water as blank control group.

As shown in Figure 3 (A), the SERS spectral curves of salbutamol using silver colloids A, B, C and D as substrates respectively showed Raman characteristic bands at 676, 809, 1061, 1157, 1250, 1299, 1350, 1469 and 1591 cm^{-1}. In addition, silver colloid D had characteristic peaks at 930 cm^{-1}. The enhanced Raman spectra of water by different colloids as control group were shown in Figure 3(B), which reflected the characteristics of colloids themself. The peak of silver colloids A, B, C and D were at 676 and 1061 cm^{-1}. In addition, silver colloid A had peak at 809 cm^{-1}, and silver colloid D had bands at 809, 930 and 1302 cm^{-1}. The SERS spectral curves of salbutamol used gold colloids F, G and H as reinforced substrates respectively had the same Raman characteristic bands. We observed that the characteristic bands existed at 801, 1299, 1373, 1548 and 2121 cm^{-1}, among which 1373, 1548 and 2121 cm^{-1} were the characteristics of gold colloids themself and only 801 and 1299 cm^{-1} were characteristic bands associated with salbutamol. The correlated bands and attribution were shown in Table 1.

Comparing different silver and gold colloids, better SERS effect and more bands in spectra were found using silver colloids as substrates for detection of salbutamol. The highest peaks occurred on enhanced Raman spectrum of salbutamol with silver colloid A as an enhanced substrate and 1157, 1250, 1299, 1350, 1469, 1591 cm^{-1} were characteristic bands associated with salbutamol. The peak at 1157 cm^{-1} is attributed to the
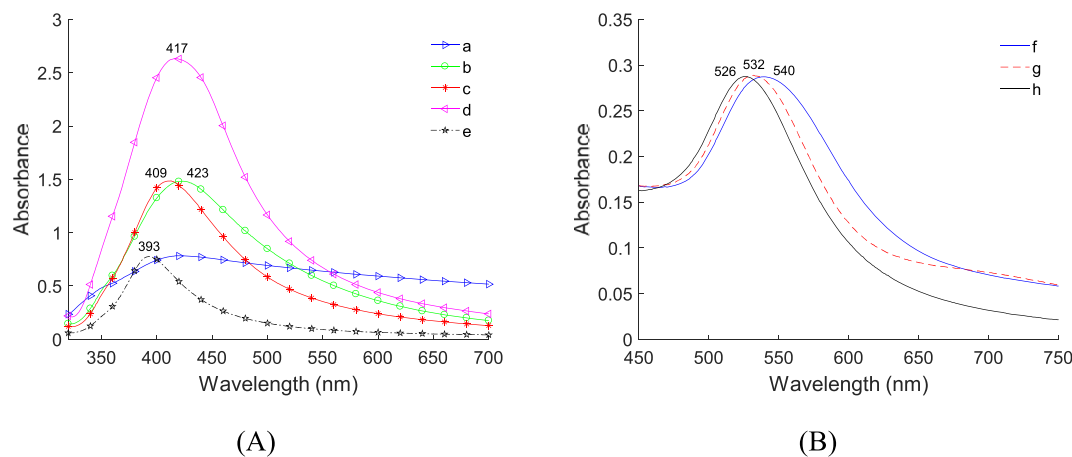

Figure 1. (A) UV-vis absorption spectra of different silver colloids: Curves a, b, c, d and e were the UV/vis absorption spectra of silver colloids A, B, C, D and E respectively; (B) UV-vis absorption spectra of different gold colloids: Curves f, g and h were the UV/vis absorption spectra of gold colloids F, G and H respectively.
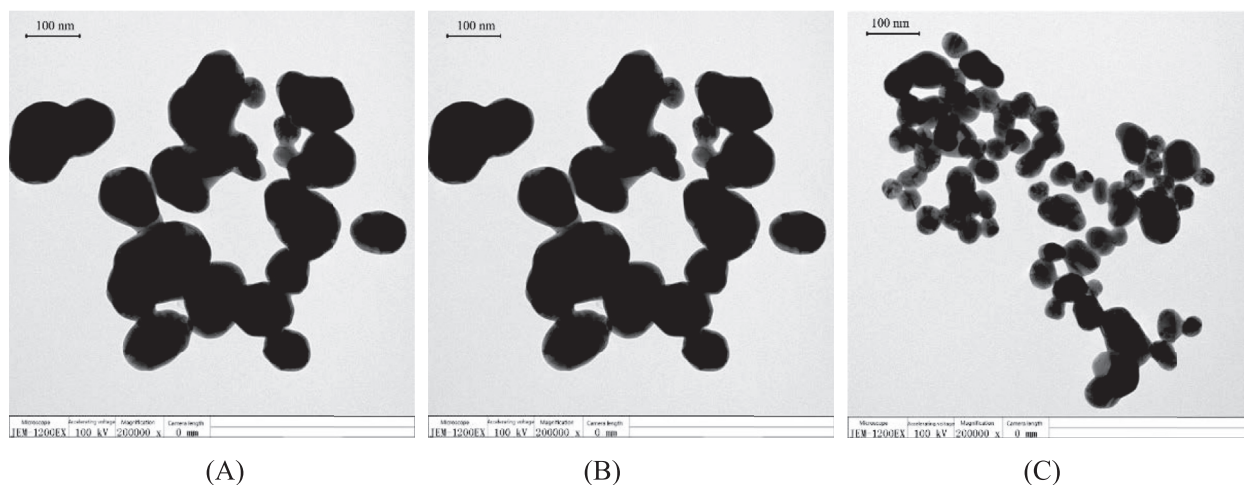
Figure 2. (A) TEM images of silver colloid A (B) TEM images of silver colloid B; (C) TEM images of silver colloid C.
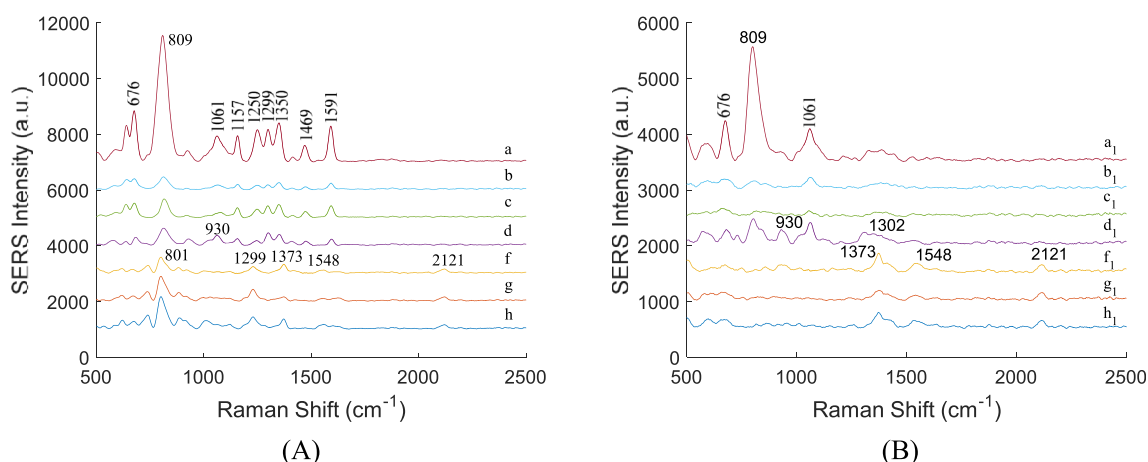

Figure 3. (A) Enhanced Raman spectra of salbutamol with different substrates: Curves a, b, c, d, f, g, and h represent Enhanced Raman spectra of salbutamol with silver colloids A, B, C, D, F, G, and H as substrate, respectively; (B) Enhanced Raman spectra of water with different enhanced substrates as a control: Curves a, b, c, d, f, g, and h represent enhanced Raman spectra of water with silver colloids A, B, C, D, F, G, and H as substrate, respectively.

Table 1. The observed Raman bands of SERS spectra and assignment.

Raman band (cm^{-1})	Proposed assignment
676, 809, 1061	Silver colloids A, B and C
676, 809, 1061, 930, 1302	Silver colloid D
1373, 1548, 2121	Gold colloids F, G and H
1157	ν(S=O)
1250	δ (C–H)
1299	ν(C–C) of the t- Butyl group
1350	t (CH$_2$), δ (C–H) and δ (O–H)
1469	δ (CH), δ (CH$_2$) and δ (CH$_3$)
1591	ν(C=C)

ν, stretching; δ, bending; t, twisting.

stretching vibration of S=O bond; The bending vibration of C–H bond is at 1250 cm^{-1}; The band centered at 1299 cm^{-1}, assigns to the stretching vibration of C–C bond in tert-butyl group; The peak at 1350 cm^{-1} is linked to the twisting of CH$_2$ coupled to C–H and O–H bending vibration (Izquierdo-Lorenzo et al., 2010); The peak at 1469 cm^{-1} is caused by bending vibrations of CH, CH$_2$ and CH$_3$ (Ali et al., 2009); The stretching vibration of C=C bond in benzene ring is observed at around 1591 cm^{-1} (Cheng et al., 2018).

Enhanced Raman spectra of 10 mg/L salbutamol solution were collected with substrates of silver colloids A, B and C respectively to explore the influence of different silver nanoparticles diameters on the adsorption effect of salbutamol. The silver colloids A, B and C were prepared by reducing silver nitrate with hydroxylamine hydrochloride and sodium hydroxide, all of which had the different average particle sizes. Silver colloids B and C were mono-dispersive and silver colloid A had polydispersity. The average particle size of silver colloid was A > B > C. The SERS peaks intensity of 10 mg/L salbutamol solution at 1157, 1250, 1299, 1350, 1469 and 1591 cm^{-1} with silver colloids A~C as substrates were compared respectively, shown in Figure 4. At the same peak, the intensity of peak was A > B > C. SERS signals are mainly generated by the target molecules adsorbed into SERS hot spots which indicates the more target molecules adsorbed into the "hot spot", the stronger the Raman signal (Etchegoin et al., 2006). The resulting particle size in the suspensions is determined by the number of nuclei over which the available silver is divided (Frens & Z., 1972). The amount of silver nitrate, reducing agent of hydroxylamine hydrochloride and sodium hydroxide added in the synthetic process of silver colloids A~C were the same. The larger the size of silver nanoparticles was, the smaller the number of nanoparticles. In this report, the average particle size of silver colloid was A > B > C, therefore, under the same volume, the concentration of silver colloids A~C nanoparticles was C > B > A. Although, the larger average particle size will reduce the number of "hot spots"
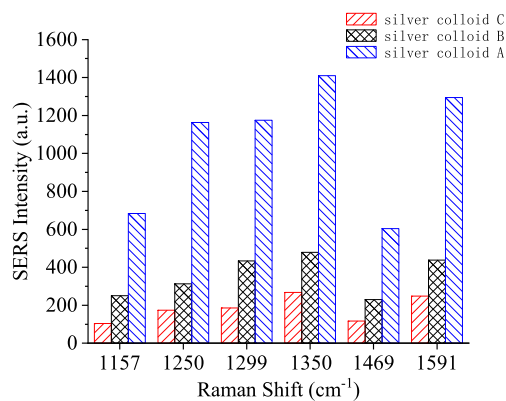
Figure 4. The SERS intensity of salbutamol solution at different peaks with silver colloids A~C as substrates respectively.

generated, there were more salbutamol molecules at the electromagnetic hot spots than silver colloids B and C which could be more easily detected by Raman probes. So, particles with larger particle size were more conducive to the adsorption of salbutamol.

3.3. Effect of different concentration of aggregating compounds on enhancement Raman spectra

Due to the difference of charge properties and binding degree between metal nanoparticles and different molecules, the adsorption degree leads to the difference of enhancement performance, which brings problems to the analysis of low concentration materials by Raman spectroscopy. The aggregating compound directly controls the enhancement of SERS signals from the analyte through the strong electric field gradient generated at the particle junction, which can lead to "hot spots". The charge balance is upset when electrolytes are added, which causes particles to coalesce (Yaffe et al., 2010). Aggregation is detrimental in some cases, however, aggregating compounds such as NaCl or NaOH are used specifically with metal colloids to generate local hot spots and facilitate SERS enhancement (Fargaš ov á et al., 2015; Xu et al., 2015). It has been studied that better enhancement performance for detection of salbutamol were found in alkaline environments. So NaOH was chosen as the aggregating compound (Izquierdo-Lorenzo et al., 2010). The enhancement performance of silver colloid can be improved by adding aggregating compound of NaOH into silver colloid and salbutamol solution.

The silver colloid A prepared above was selected as the enhanced substrate, and the sodium hydroxide aqueous solution of 0.1, 0.2 and 0.3 M were selected as the aggregating compounds respectively. The SERS spectra of salbutamol solution (10 mg/L) were collected to explore the influence of different concentrations of sodium hydroxide aqueous solution on the enhancement performance shown in Figure 5 (A). Figure 5 (B) showed the SERS intensity of salbutamol solution at different peaks with different concentration of NaOH solutions as aggregating compounds. It could be seen the SERS peaks intensity of 10 mg/L salbutamol solution at 1157, 1250, 1299, 1350, 1469 and 1591 cm^{-1} were the highest with 0.2 mol/L NaOH solution as an aggregating compound.

3.4. Quantitative analysis

The SERS spectra of 0.1–10 mg/L salbutamol solution were collected by using 0.2 mol/L NaOH aqueous solution as aggregating compound and silver colloid A as enhanced substrate. Multiple scattering correction (MSC) and Automatic Whittaker Filter (AWF) were used for pre-processing to reduce fluorescence background contained in the original Raman spectra collected. The bands at 1157, 1250, 1299, 1350, 1469, 1591 cm^{-1} were associated with salbutamol and the peak intensity gradually decreased with the decrease of salbutamol solution concentration. With the concentration of salbutamol decreased to 0.5 mg/L, the SERS spectra peaks of salbutamol solution gradually disappeared at 1157 cm^{-1}, 1250 cm^{-1}, 1299 cm^{-1}, 1350 cm^{-1} and 1469 cm^{-1}, However, the peak at 1591 cm^{-1} disappeared in the SERS spectra of 0.1 mg/L salbutamol solution.

In order to improve the ability of SERS in the trace detection of salbutamol, the characteristic peak intensity of salbutamol (0.2–10 mg/L) at 1591 cm^{-1} was selected for data analysis. As shown in Figure 6(A), with the increase of salbutamol concentration, the peak intensity of salbutamol at 1591 cm^{-1} tended to increase and fluctuated within a certain range. A better linear relationship between the concentration of salbutamol (ranged from 0.2 to 1 mg/L) and SERS intensity was found in Figure 6(B). The equation of the calibration curves between Raman intensity and salbutamol concentration was shown in Formula (1).

$$C = 0.0023 \cdot I \text{-} 0.079 \tag{1}$$

Where C (mg/L) is the concentration of salbutamol solution and I is the SERS intensity of salbutamol solution. Calibration set correlation coefficient is 0.988 and prediction root mean square error is 0.029 mg/L.
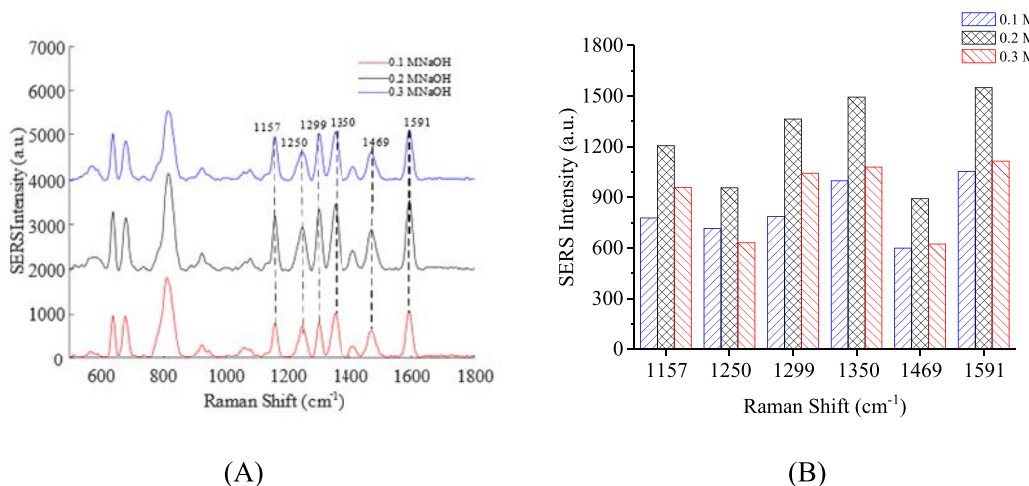

(A)

(B)

Figure 5. (A) The SERS spectra of salbutamol solution with different concentration of NaOH solutions as aggregating compounds; (B) the SERS intensity of salbutamol solution at different peaks with different concentration of NaOH solution as aggregating compounds.
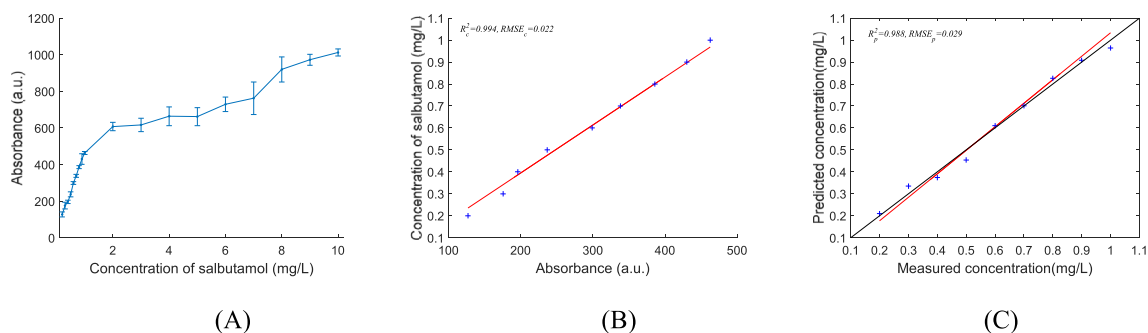
Figure 6. (A) The relationship between Raman intensity and salbutamol concentration (0.2–10 mg/L) (B) The relationship between Raman intensity and salbutamol concentration (0.2–1 mg/L); (C) The plot of the salbutamol concentration determined by SERS versus the spiked salbutamol concentration.

Nine salbutamol samples with a concentration gradient of 0.2–1 mg/L were prepared for SERS spectra detection and the Raman spectra obtained were preprocessed with MSC and AWF. The peak intensity at 1591 cm^{-1} of spectra were interpolated into the equation of the calibration curves to verify the prediction performance of the model. As shown in Figure 6(C), the model had a better performance with a good linearity (R^2 =0.988) and lower root mean square error ($RMSE_c$ = 0.029 mg/L).

SERS spectroscopy has been proven to be a suitable technique for trace analysis of salbutamol using gold colloid. In this paper, concentrated silver colloid was used as an enhanced substrate and had better predictive ability for salbutamol, with low cost and detection limit of 0.2 mg/L. Compared with previous studies, the detection limit was further reduced, however, compared with traditional liquid chromatography, the detection limit still needs to be further reduced. In this paper, the adsorption effects of silver colloid with different particle sizes on salbutamol were compared. As a result, silver nanoparticles with larger particle sizes were more conducive to the adsorption of salbutamol which made a great contribution to further reducing the detection limit of salbutamol by SERS. However, the optimal particle size for salbutamol adsorption needs to be further explored which is beneficial to improve SERS enhanced performance. With the rapid updating of Raman system hardware and the gradual deepening of stoichiometry, the detection limit will be gradually reduced.

4. Conclusion

In this report, the SERS detection of salbutamol were carried out by using several different silver colloids and gold colloids with different particle sizes as enhanced substrates. It was found that silver colloid reduced by hydroxylamine hydrochloride had the best enhancement performance on salbutamol. The adsorption effect of several silver colloids with different particle sizes on salbutamol was compared. It was found that the larger particle size of silver nanoparticles was, the better enhancement effect was, which indicated silver nanoparticles with larger particle sizes were more conducive to the adsorption of salbutamol.

Under the optimal conditions (Silver colloid A as enhanced substrate, 0.2 mol/L NaOH aqueous solution as aggregating compound), a better linear relationship between the concentration of salbutamol (ranged from 0.2 to 1 mg/L) and SERS intensity. The equation of the calibration curves was $C = 0.0023 \bullet I$-0.079 (mg/L) with a good linearity (R^2 =0.994) and lower root mean square error ($RMSE_c$ = 0.022 mg/L). Validation set correlation coefficient was 0.988 and prediction root mean square error was 0.029 mg/L. This method provides a new idea for further reducing the detection limit of salbutamol.

Declarations

Author contribution statement

Qinghui Guo, Yankun Peng: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Kuanglin Chao: Conceived and designed the experiments; Analyzed and interpreted the data.

Data availability statement

Data will be made available on request.

Declaration of interests statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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