A novel Ag/AgO/carboxymethyl chitosan bacteriostatic hydrogel for drug delivery

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Abstract
pH-sensitive Ag/AgO/carboxymethyl chitosan (CMCS) bacteriostatic hydrogels and Ag/AgO/CMCS/Aspirin (ASP) carrier hydrogels were prepared by Ag/AgO in situ precipitation method, and the effects of swelling, degradation, drug release and antibacterial properties of hydrogels were studied. The network of Ag/AgO/CMCS/ASP drug-loaded gels produced was mainly cross-linking by hydrogen bonding and intermolecular forces, and the cross-linking silver was mainly present in the elemental Ag and Ag2+ states. Under the condition of buffer solution pH = 7.4, the cumulative release amount of Ag/AgO/CMCS/ASP drug-loaded gel was 75.20% within 12 h, and the inhibition rate of Gram-negative Escherichia coli (E. coli) reached the maximum of 92.32%, which had broad application prospects in the medical field.

1. Introduction

With the rapid development of drug treatment, the concept of ‘optimization of medication’ has gradually become a hot spot of concern. If the administration process can accurately match the physiological requirements at the appropriate time or at the appropriate place, it can effectively improve the bioavailability of the target site and reduce the side effects [1]. As soft biomaterials, hydrogels have been used extensively in the area of tissue engineering and drug delivery because of their three dimensional networks, high water content, flexibility, good biocompatibility and adaptive biodegradability [2–7]. Hydrogels can be classified into smart hydrogels and ordinary hydrogels according to their responsiveness to external environmental conditions. Intelligent hydrogels have important research significance by reacting to external environmental factors such as temperature [8], light [9], magnetic field [10], electric field [11], pH [12–14], etc. A number of hydrogels based on natural materials or synthetic polymers have been developed and served as drug carriers since they can absorb a large amount of water-soluble drugs in their three-dimensional polymeric networks [15–17].

Previous studies on hydrogel systems had focused on type of crosslinking agent [18] and the effect of new synthetic methods [19] on the gelation time, mechanical properties, micromorphology or drug release of hydrogels. Due to the superiority of biomedicine, developing antibacterial hydrogels is becoming more practical. Wahid et al [20] successfully prepared carboxymethyl chitosan (CMCS)/ZnO nanocomposite antibacterial hydrogel by in situ synthesis of zinc oxide (ZnO) nanorods. Zhou et al [21] synthesized nano-silver/gelatin/carboxymethyl chitosan antibacterial hydrogel by radiation-induced reduction at room temperature. Marek et al [22] synthesized chitosan–silver hydrogels used for modification of cotton fabric in order to give it antimicrobial properties. Mehdii et al [23] prepared a novel chitosan/silver nanocomposite hydrogel (CH/AgNPs) by in situ formation of AgNPs in the chitosan hydrogel matrix.

However, there have been few reports on the preparation of antibacterial hydrogels by applying the antibacterial properties of silver ions (Ag2+). Silver and silver ions have been known as effective antimicrobial agents for a long time. Nanosilver particles have extremely large surface area to contact with bacteria or fungi.
They could bind to microbial DNA and the sulphhydryl groups of the metabolic enzymes in bacterial electron transport chain. The former will prevent the replication of bacteria, and the latter will inactivate them [24, 25]. Hence, owing to their excellent antimicrobial activity, nanosilver particles have been applied to a wide range of healthcare products such as burn dressings, scaffolds, skin doners and medical devices [26, 27].

In this study, carboxymethyl chitosan (CMCS) as an initial material, which is water-soluble, non-toxic, biodegradable and biocompatible, and has been extensively investigated in applications of pharmaceuticals and biomedical materials [7, 28–30], silver nitrate as crosslinking agent, pH-sensitive Ag/AgO/CMCS antibacterial hydrogels were prepared via an in situ precipitation method in a mild condition. Fourier transform infrared (FTIR), X-ray diffraction (XRD), X-ray photoelectron spectroscopy (XPS), Scanning electron microscope (SEM) and other characterization methods were used to study the physical properties of the hydrogel such as the pore structure, cross-linking mechanism, and thermal stability. The swelling kinetics, the effects of pH on swelling, degradation, and aspirin drug release behavior were investigated. In addition, the antibacterial properties of hydrogels against Gram-negative Escherichia coli were tested. Ag/AgO/CMCS hydrogel combines the advantages of both CMCS and Ag/AgO, and is expected to become a new type of antibacterial hydrogel.

2. Experimental

2.1. Materials

Carboxymethyl chitosan (CMCS, Biological reagent) was purchased from Shanghai Aladdin Chemistry Co., Ltd. Gelatin was obtained from Chengdu Chemical Reagent Corporation (Chengdu, China). Aspirin (ASP) was acquired from Chongqing Boyi Chemical Reagent Corporation (Chongqing, China). Silver nitrate (AgNO3) and ammonium hydroxide (NH4OH) were analytical grade products obtained from Sinopharm Chemical Reagent Co., Ltd. Escherichia coli (E. coli) All other reagents were of analytical grade and used as received.

2.2. Instrumentation

Fourier transform infrared (FTIR) spectra were recorded on a Nicolet Magna IR 550 spectrometer in KBr pellets. X-ray photoelectron spectroscopy (XPS) spectrum was performed on a ThermoFisher Scientific ES-CALAB 250Xi spectrometer. A Shimadzu UV-visible spectrophotometer (DUV-3700) was used to record the absorbance of samples. X-ray diffraction (XRD) measurements were conducted with solid samples using a PANalytical X’pert Pro MPD (40 kV, 40 mA). Solid samples were mounted on a sample holder and scanned from 2θ = 5 to 80° at a speed of 4° min⁻¹. Scanning electron microscope (SEM) images were obtained using a ThermoFisher Scientific Scanning Electron Microscope (JSM-64901LV). Thermograms of samples were recorded on a Shimadzu Differential Thermal Thermogravimetric Analyzer (DTG-60H).

2.3. Preparation of samples

CMCS (1.2 g) and gelatin (1.2 g) were dissolved in 20 ml of deionized water at 70 °C, and the mixture was stirred for 1 h until the two were mixed well. The ammonium hydroxide solution (1 ml, 0.1 mol l⁻¹) added with a certain amount of AgNO3 was added dropwise to the mixed solution. The mixed solution was left at room temperature for 24 h, frozen, and dried to obtain Ag/AgO/CMCS hydrogels. According to the increasing concentration of the cross-linking agent AgNO3, Ag/AgO/CMCS hydrogels were recorded as Ag/AgO/CMCS-1, Ag/AgO/CMCS-2, Ag/AgO/CMCS-3, Ag/AgO/CMCS-4, Ag/AgO/CMCS-5 in sequence.

0.1 mg of ASP was dissolved in 20 ml of mixed solution of gelatin and CMCS. Ag/AgO/CMCS ASP drug-loaded hydrogels were obtained using the same method as above. According to the increasing concentration of the cross-linking agent AgNO3, Ag/AgO/CMCS hydrogels were recorded as Ag/AgO/CMCS/ASP-1, Ag/AgO/CMCS/ASP-2, Ag/AgO/CMCS/ASP-3, Ag/AgO/CMCS/ASP-4, Ag/AgO/CMCS/ASP-5 in sequence.

2.4. Sample testing

2.4.1. Swelling ratio of Ag/AgO/CMCS hydrogel

Two 0.2 g of frozen and dried Ag/AgO/CMCS hydrogels were placed in 50 ml of buffer solution simulating gastric and intestinal fluids respectively at 37 °C. The hydrogels were taken at regular intervals, and the surface moisture was dried and weighed. Measure three times in parallel, take the statistical average, and insert it into the swelling ratio (SR) calculation formula as follows:

\[ \text{SR} = \left( \frac{W_t - W_0}{W_0} \right) \times 100\% \]

Among them, \( W_0 \) is the initial mass of the hydrogel, g; and \( W_t \) is the mass after swelling t time, g.
2.4.2. Degradation rate of Ag/AgO/CMCS hydrogel

A certain amount of hydrogel (0.2 g) were put into 100 ml of buffer solution simulating gastric and intestinal fluid, taken out at a certain interval, dried and weighed. Measure three times in parallel, take the statistical average, and insert it into the degradation rate (DR) calculation formula as follows:

\[
DR = \frac{W_d - W_i}{W_d} \times 100\%
\]

Among them, \(W_d\) is the initial mass of the hydrogels, g; and \(W_i\) is the mass after swelling i time, g.

2.4.3. Determination of Aspirin (ASP) drug release

0.2 g of Ag/AgO/CMCS ASP drug-loaded hydrogels were placed in 100 ml of a buffer solution of pH 7.4 and pH 2.0 with a concentration of 0.1 M, and ASP drug was slowly released in a constant temperature shaker at a shaking rate of 50 rpm at 37 °C. 4 ml of mixture was taken out at a certain interval and fresh original buffer solution was replenished. The absorbance at 222 nm was measured with an ultraviolet-visible spectrophotometer at 37 °C, and measured in parallel three times, and the statistical average was taken. The cumulative release (CR) calculation formula as follows:

\[
CR = \frac{m_n}{m_0 \times 0.1} \times 100\%
\]

\(C_n\) is the mass concentration of ASP in the nth sampling, mg l\(^{-1}\); \(m_n\) is the cumulative release mass of the ASP in the nth sampling, mg; and \(m_0\) is the initial mass of hydrogel, mg.

2.4.4. Determination of antibacterial performance of Ag/AgO/CMCS hydrogels

- Bacteriostatic zone test

\(E. coli\) was evenly spread on a nutrient agar plate medium at 37 °C, and three hydrogel cylindrical slices (Ag/AgO/CMCS-0, 1, 5, d = 10 mm, h = 5 mm) were placed on the medium and incubated for 12 h. The bacterial growth around the sheet was observed and the diameter of the zone of inhibition was measured.

- Bacterial inhibition kinetic curve

0.5 g of hydrogels were weighed and immersed in 40 ml of \(E. coli\) (OD = 1) nutrient agar culture medium, and blank \(E. coli\) medium was used as a control. The sample was incubated in a thermostatic shaker for 12 h at a shaking rate of 50 rpm at 37 °C. At a certain time interval, 3 ml of the culture solution was taken out, and the absorbance of the culture solution at 600 nm was measured with a UV-visible spectrophotometer. The inhibition ratio (IR) calculation formula is as follows:

\[
IR = \left(1 - \frac{A_t - A_0}{A_{con} - A_0}\right) \times 100\%
\]

In the formula, \(A_0\) is the absorbance before incubation of \(E. coli\); \(A_t\) and \(A_{con}\) are the absorbance of \(E. coli\) in the hydrogel and the reference sample after incubation for t time.

3. Results and discussion

3.1. Fourier transform infrared (FTIR) analysis

The FTIR spectra of carboxymethyl chitosan, gelatin and hydrogel are shown in figure 1. In carboxymethyl chitosan, the absorption peak at 3438 cm\(^{-1}\) is the result of overlapping stretching vibrations of \(-\text{NH}\) and \(-\text{OH}\), the stretching vibration peak at 1737 cm\(^{-1}\) is C=O. The absorption peak of 1634 cm\(^{-1}\) is the result of the \(-\text{NH}_2\) deformation vibration and \(-\text{COO}^-\)– asymmetric stretching vibration overlapping. The absorption peak at 1386 cm\(^{-1}\) is \(-\text{COO}^-\)– symmetrical stretching vibration. The stretching vibration absorption peaks of C–N and C–O are at 1112 cm\(^{-1}\) and 1023 cm\(^{-1}\), respectively. The characteristic absorption peaks at 3445 cm\(^{-1}\), 1742 cm\(^{-1}\), 1641 cm\(^{-1}\), and 1399 cm\(^{-1}\) in gelatin are \(-\text{NH}\) and \(-\text{OH}\) stretching vibration, C=O stretching vibration, \(-\text{NH}_2\) deformation vibration, \(-\text{COO}^-\)– result of symmetrical stretching vibration. Comparing the FTIR spectra of hydrogel with carboxymethyl chitosan and gelatin monomers, there is no significant difference overall, but the stretching vibration peaks of \(-\text{NH}_2\), \(-\text{OH}\) and \(-\text{COO}^-\)– are significantly shifted to low wave numbers, indicating that there is a strong intermolecular hydrogen bonding between carboxymethyl chitosan and gelatin. At the same time, the symmetrical stretching vibration peak of \(-\text{COO}^-\) at 1386 cm\(^{-1}\) shifted to a
high wave number, which further proved that there is a strong intermolecular interaction between the two molecules.

3.2. X-ray diffraction (XRD) analysis
Figure 2 shows the XRD spectra of CMCS, gelatin and Ag/AgO/CMCS hydrogel. At $2\theta = 8^\circ$ and $21.5^\circ$, the three samples all show obvious doublet, which is the reflection crystal plane caused by the superposition between aromatic layers. $2\theta = 38^\circ, 44^\circ, 64^\circ$, and $78^\circ$ are the reflective crystal planes (111), (200), (220), and (311) of metallic Ag, respectively. The peak at $2\theta = 32^\circ$ is the (111) crystal plane of AgO. These results indicate the coexistence of Ag and AgO phases in the hydrogel samples [31].

3.3. X-ray photoelectron spectroscopy (XPS) analysis
In order to further determine the valence state of silver in the hydrogel, a sample of Ag/AgO/CMCS-3 with a moderate concentration of AgNO$_3$ was selected for X-ray photoelectron spectroscopy analysis. The XPS spectrum of the Ag/AgO/CMCS-3 hydrogel is shown in figure 3. Ag/AgO/Ag CMCS-3 d of XPS spectra of the sample is fitted to two superimposed doublet (figure 3(a)). The peaks at 367.8 and 373.9 eV are elemental Ag, and the peaks observed at 367.4 eV and 373.4 eV are Ag$^{2+}$, which are consistent with the XRD results. The C 1 s XPS spectrum is shown in figure 3(b). Four peaks were separated by fitting. The peak at the binding energy value of 284.2 eV is due to the existence of CN structure in the skeleton. The peaks at 284.7 eV and 285.9 eV correspond to CO and OCO structure, corresponding to $-\text{COO}^-$ structure at 287.6 eV. In the N 1 s spectrum (figure 3(c)), because its N element mainly exists in the C-NH$_2$ state and is affected by the macrocyclic structure and the long molecular chain, its corresponding N 1s binding energy is at 399.0 eV without deacylation. The structure of the chemical moiety ($-\text{NHCOCCH}_3$) decreases the electron cloud density around the N atom due to the electron-drawing effect of $-\text{COCH}_3$, and the binding energy moves toward 399.7 eV. In the O 1 s spectrum (figure 3(d)), the peak near 530.8 eV is AgO, and the second peak at 531.7 eV is oxygen on the polymer segment.

3.4. Scanning electron microscope (SEM) analysis
The surface morphology SEM of the hydrogel is shown in figure 4. A continuous porous three-dimensional network structure with a smooth surface is formed in the hydrogel, and the average pore size distribution is in the range of 5–10 $\mu$m. This porous structure allows water molecules to enter the hydrogel network, a region of water seepage and interaction sites in biological media or buffers. The prepared silver and silver oxide particles are not visible in the SEM image because they are relatively small in size.
3.5. Thermal gravimetric analyzer (TGA) analysis
As shown in figure 5, the weight loss of all monomers and hydrogels can be divided into three stages in the temperature range of 25 to 500 °C. The weight loss in the first stage is the loss of free water molecules in the...
monomer and the hydrogel. It is observed that the difference in weight loss of each component at this stage is small, and the residual weight is about 80%. With the temperature rising gradually, the temperature range of about 234 °C ∼ 392 °C, the monomer and hydrogel in turn enter the second stage of weight loss process, this stage is mainly due to the decomposition of functional groups on the polymer chain to promote weight reduction, each degree of reduction difference. Functional groups such as carboxyl groups on the gelatin skeleton are thermally decomposed, resulting in a rapid decrease in the weight of the gelatin, with a residual weight of 33.02% at 392 °C. The carboxymethyl group in carboxymethyl chitosan was decomposed, and the residual weight was 46.17% at 392 °C. Due to the formation of the crosslinked structure in the drug-loaded hydrogel and blank hydrogel, the weight loss during the second stage of degradation was smaller than that of the monomer, and the residual weights were 49.12% and 45.84% at 392 °C, respectively. In the temperature range of about 392 °C ∼ 500 °C, the skeleton of monomer and hydrogel gradually begins to degrade under high temperature. It can be seen from the figure that the residual weight value of the drug-loaded hydrogel is higher than that of the monomer and the drug-free hydrogel, indicating that the thermal stability of the hydrogel is higher than the thermal stability of each monomer. The glue has higher thermal stability due to the drug. This higher thermal stability is due to the strong bonding due to the interaction force between the substances.
3.6. Swelling and degradation properties of Ag/AgO/CMCS hydrogel

The average swelling rates of all hydrogel preparations (Ag/AgO/CMCS-1~5) are shown in figure 6(a). When the pH $< 7.4$, the swelling rate of the hydrogel is proportional to the pH value, and the swelling rate reaches the maximum value when the pH is 7.4. As the pH continues to increase, the swelling rate begins to decrease. This is because when the pH value in the medium changes, the hydrophilic properties of the free amino groups and carboxyl groups on the carboxymethyl chitosan and gelatin molecular chains will change significantly, thus showing different swelling properties. When pH $< 7.4$, the carboxylated chitosan and the $-\text{COOH}$ moiety on the gelatin chain were ionized and existed as $-\text{COO}^-$. The electrostatic repulsion and hydrophilicity between $-\text{COO}^-$ cause the gel network to swell, thereby increasing the pores and free space within the hydrogel and absorbing more water accordingly. At pH $= 7.4$, $-\text{COOH}$ is completely ionized into $-\text{COO}^-$ form, and $-\text{NH}_3^+$ is also deprotonated to $-\text{NH}_2$, and the strong static repulsion between $-\text{COO}^-$ and hydrophilicity make the hydrogel swelling rate reach the maximum. The pH continued to increase, and $-\text{NH}_3^+$ was completely deprotonated, resulting in a decrease in the solubility of the molecular chain and the formation of new hydrogen bonds, which caused the polymer chain to contract, the swelling rate decreased. Therefore, as the pH value

Figure 6. (a) Swelling equilibrium curves of Ag/AgO/CMCS gels in different pH buffers (b) Ag/AgO/CMCS gel swelling curves in pH 7.4 buffer solutions.
increases in the alkaline environment, the swelling degree decreases. As shown in figure 6(b), at pH = 7.4, as the concentration of AgNO₃ increases, the hydrogel exhibits a slower swelling rate and a smaller swelling degree, indicating that there is more cross-linking point in the hydrogel network, preventing the aqueous medium from entering the hydrogel \[32, 33\].

The degradation rate of Ag/AgO/CMCS-3 in different pH media is shown in figure 7(a). Similar to the swelling behavior, the cross-linked structure of the hydrogel was effectively destroyed due to the electrostatic repulsion in pH 7.4 medium, and its degradation rate was faster than under weakly acidic or alkaline conditions, with a degradation rate of 92% in 18 days. In the acidic medium, Ag/AgO/CMCS-3 hydrogel hardly degrades, indicating that the hydrogel can keep the intact state when it enters the gastric juice. In addition, as shown in figure 7(b), in the pH 7.4 medium, the larger the AgNO₃ concentration in the hydrogel, the lower the degradation rate, indicating that the crosslinking density of the hydrogel is increased, and the degradation of the hydrogel in the medium is suppressed effect.
3.7. Controlled release properties of Ag/AgO/CMCS hydrogels

Figure 8(a) is the drug release behavior of ASP drug-loaded hydrogel (Ag/AgO/CMCS/ASP-1~5) in simulated intestinal fluid. It was found that the higher the AgNO₃ concentration, the lower the drug release rate and the smaller the drug release amount. This is because the concentration of AgNO₃ increases, the crosslink density of the hydrogel network becomes larger, the space and pore channels provided for drug loading and diffusion become smaller, and the tight crosslink network hinders the swelling and degradation behavior of the hydrogel. The release rate and the release amount are reduced. Figure 8(b) shows the release behavior of Ag/AgO/CMCS/ASP-5 drug-loaded hydrogel in simulated gastric and intestinal fluid. ASP has a total release of 75.20% in simulated intestinal fluid for 12 h and 20.14% in simulated gastric fluid. The amount of drug released in the intestinal fluid environment is significantly higher than that in the gastric fluid environment, which is consistent with the swelling and degradation behavior of the gel at different pH conditions. The relatively loose structure produced after swelling or degradation reduces the resistance to drug diffusion, so the drug release rate is accelerated.
3.8. Release kinetics of Ag/AgO/CMCS hydrogel

To understand the release mechanism of encapsulated drugs, we fit the cumulative drug release data using the Ritger–Peppas equation as shown below:

\[
\frac{M_t}{M_\infty} = k t^n \left( \frac{M_t}{M_\infty} \right)^{0.6}
\]

In the formula, \(M_t\) and \(M_\infty\) are the cumulative drug release at time \(t\) and equilibrium, respectively, \(k\) is a rate constant related to the hydrogel matrix and the properties of the drug, and \(n\) is a release index characterizing the drug transport mechanism. If \(n \leq 0.45\), it corresponds to Case I diffusion behavior and follows the Fickian diffusion mechanism. When the value of \(n\) is between 0.45 and 0.89, it corresponds to the irregular drug delivery behavior of Case III, following the Non-Fickian diffusion mechanism, and the drug release is affected by the synergy of diffusion and erosion. If \(n > 0.89\), it corresponds to Case II swelling controlled release behavior, similar to the zero order release mechanism.

Table 1 lists the corresponding drug release kinetics data obtained by fitting the drug release experimental data to the Ritger–Peppas equation. When \(pH = 7.4\), the release index \(n\) is between 0.45 and 0.89, following the Non-Fickian diffusion mechanism, which shows that ASP drug release is affected by the synergistic effect of diffusion and erosion. When \(pH = 1.2\), the release index \(n = 0.351 72 < 0.45\), ASP drug release follows the Fickian diffusion mechanism, and hydrogels mainly swell in the acidic environment to release drugs, which is consistent with the analysis above. The crosslinking density of hydrogel has little effect on drug release kinetics and does not change ASP release mechanism.

3.9. Antibacterial properties of Ag/AgO/CMCS hydrogel

As shown in figure 9(a), the diameters of the inhibition rings of the Ag/AgO/CMCS-1 and Ag/AgO/CMCS-5 hydrogels were 11.8 and 13.6 cm, respectively. As a contrast, no obvious inhibition circle was found in the Ag/AgO/CMCS-0 hydrogel. Therefore, the antibacterial activity is mainly attributed to the presence of Ag/AgO particles in the hydrogel. The bacteriostatic ability of the Ag/AgO/CMCS hydrogel is expressed by the inhibition rate. As shown in figure 9(b), it was found that the inhibition rate of Ag/AgO/CMCS-5 on E. coli increased by multiples within 6 h of incubation, and the maximum inhibition rate was 92%. Within 12 h of incubation, the inhibition rate remained above 90%. Subsequently, the suppression rate gradually decreased to zero. The maximum inhibition rate of Ag/AgO/CMCS-1 hydrogel on E. coli was 37%. This is because when the hydrogel is immersed in the suspension, a large number of Ag/AgO particles are released and contribute to the initial inhibition of bacterial growth. At the same time, these particles interact with the disrupted intracellular material.
and form coagulum, leading to a reduction in Ag/AgO concentration. As the release of Ag/AgO gradually decreases, the consumption of Ag/AgO is dominant. Therefore, when Ag/AgO concentration falls below the minimum inhibitory concentration, bacteria will resume growth. However, it was observed that Ag/AgO/CMCS-0 still had a weak bacteriostatic effect. This is because carboxylated chitosan itself has a certain degree of antibacterial ability.

4. Conclusion

Using AgNO₃ as a cross-linking agent, Ag/AgO/carboxymethyl chitosan hydrogels with different cross-linking densities were synthesized. The structure and composition of the hydrogel, swelling kinetics, in vitro degradation characteristics, and sustained-release properties of the drug were studied. The research and evaluation of the antibacterial performance with *Escherichia coli*, the following conclusions were obtained:

The structure and composition of the hydrogel were characterized by FTIR, XRD and XPS. The hydrogel network was analyzed by physical cross-linking, and the cross-linked silver was mainly in the elemental Ag and Ag⁺⁺ states.

The residual weight of the drug-loaded hydrogel was higher than that of the monomer and the drug-free hydrogel, and the residual weight of 468.82 °C was 43.05%. It can be seen that the thermal stability of the hydrogel is higher than that of each monomer. The swelling and degradation behaviors of Ag/AgO/carboxymethyl chitosan hydrogels are similar. The swelling degree and degradation rate increase first and then decrease in the pH range, reaching a maximum value at pH = 7.4. At the same time, increasing the crosslinking density has an inhibitory effect on them.

The total drug release of Ag/AgO/CMCS/ASP-5 drug-loaded gel in simulated intestinal fluid for 12 h was 75.20% and 20.14% in simulated gastric fluid. The amount of drug released in the intestinal fluid environment was significantly higher than that in the gastric fluid environment.

The ASP drug-loaded gel follows the Non-Fickian diffusion mechanism at pH = 7.4 and the Fickian diffusion mechanism at pH = 1.2.

The inhibition rate of Ag/AgO/CMCS-5 on *Escherichia coli* was 92%. The results show that the hydrogel has the advantages of promoting the cumulative release of ASP in the intestinal site and improving the bacteriostatic performance, is a good and effective method of drug administration, and has broad application prospects in the treatment of pain.

Notes

The authors declare no competing financial interest.

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