Sterilization effects of several metal nanoparticles

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Abstract: The luminescence characteristics of Escherichia coli (E. coli) were studied by fluorescence spectroscopy technique. The fluorescent method was used to characterize the sterilization effect of three nanometals on E. coli. The experimental results show that E. coli has two characteristic emission peaks; The fluorescence characteristics of E. coli were used to characterize the effect of silver nanoparticles on the sterilization of E. coli at different time, dosage and temperature. The experimental results showed that the gold nanoparticles are more suitable for sterilization. The results of this study have reference significance for the count and antibacterial study of E. coli in environment, food and so on.

Keyword: Nanometal; E.coli; Sterilization.

I. INTRODUCTION

Escherichia coli also called colon bacillus, is a flagella, can exercise, spores-free prokaryotes, the structure is simple and easy to cultivate, the plasmid in the cytoplasm are often used as the object of genetic engineering. 1885, Escherich discovered E. coli parasites in the intestine, and for a long time thereafter, E. coli was considered as one of the intestinal bacteria. Under normal circumstances, E. coli is a probiotics of the human body, can competitively resist pathogens attack, and can the synthesize vitamin B and K, and symbiosis with human body. In the middle of the 20th century, Europe and other countries issued bloody diarrhea, Riley and Remis [1] et al. after the investigation of the cause of the disease, found that E. coli O157:H7, thus sparked a hot research on E. coli. Today, pathogenic E. coli caused by food poisoning events be too numerous to enumerate [2, 3].

Nanometal particles are most commonly used for antimicrobial purposes, because of their very strong bactericidal ability, make very small amount of silver nanoparticles can kill Hundreds kinds of bacteria in a few minutes, broad-spectrum sterilization without any resistance, toxicity small reaction, almost no irritation, is the latest generation of natural antibacterial agents. The excellent antibacterial ability of silver nanoparticles has led many people to study.

Most of the microbial counting for the traditional plate count method, this method takes a long time, and the operation method is relatively complicated. Fluorescence spectrum detection method has the characteristics of high sensitivity, high speed and strong stability [4]. In the paper the fluorescence spectrum detection technology has been used to study the fluorescence characteristics of E. coli, and the number of E. coli was characterized by its emission peaks, makes it easier to count E. coli.

II. EXPERIMENTAL EQUIPMENT AND SAMPLES

A. Instruments and parameters

The experiment equipment was FLS900 multifunctional spectrometer produced by Edinburgh Instruments Company. The samples emission spectra were scanned within in the range of 310~550 nm. The excited light source was a xenon lamp with the output wavelength of 289 nm.

B. Main reagent

E. coli (solid, ATCC25922 standard strain, stored at 5 C), LB liquid medium (dry powder, Tryptone 10g, Yeast Extract 5g, NaCl (sodium chloride) 5g).

C. Sample preparation

1) The liquid strain preparation: that is, a generation of E. coli. In a sterile environment, E. coli (solid) bacteria inoculated into prepared sterile culture medium, and the E. coli solution was shake culture at 37 ℃ for 24h with a shaker (120rpm). Take 2 ml of E. coli solution, centrifuged centrifuge for 10 minutes at 2000rmp, with a sterile pipette sucked the liquid above. Take 0.024g of E. coli, add 2 ml sterile culture dilution, shake, as the second generation of E. coli strains and spare.

2) The second generation E. coli preparation: Take 500 μ L of the first generation E. coli solution, added into 50 ml of sterile culture solution, shook for 15h at 37 ℃ environment (120rpm). Take out 2 ml of the cultured second generation E. coli solution, put into a 4 ml centrifuge tube, centrifuge for 10 minutes at 2000rpm , aspirate the above liquid, add 4 ml and physiological saline, and repeat 3 times, to make the required experimental samples. As the second generation E. coli solution by adding 4ml physiological saline, formulated into E. coli solution for use.

In the sample pretreatment, the first generation of E. coli is used as the strain, mainly to ensure the second generation of E.coli culture conditions of consistency, try to avoid the experimental error.

III. Results and Discussion

A. Relationship between fluorescence intensity and concentration of E. coli

In this experiment, 2 ml of the second generation of E. coli cultured for 24 hours was taken out, centrifuged for 3 times and then added with 4ml of physiological saline to shake to prepare the solution of E. coli. The solution was irradiated with 289 nm excitation light to obtain the emission spectrum of Escherichia coli in figure 1. It can be seen from the figure that E. coli has a very wide emission peak near 332 nm and a less intense emission peak near 425 nm. The emission peak near 332 nm is caused by the superposition of amino acid tyrosine and tryptophan in protein [5]. The emission peak near 425 nm is generated by the superposition of nucleic acid and NADH [6].

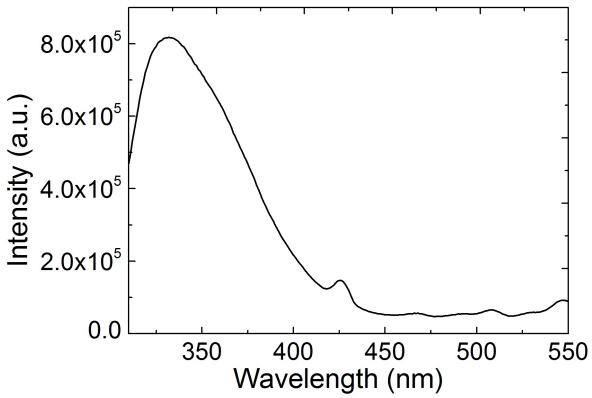

Fig.1. Emission spectra of E. coli

B. Comparison of the sterilization effects of several nano-metals

1) Speed of sterilization

Take out the prepared E. coli solution and add 0.5 ml Nano Silver, Nano Gold and Titanium Dioxide respectively and placed in an environment at 30 □. The fluorescence spectra of sample in the range of 310 nm ~ 550 nm were measured every 1 hour. The intensity of the characteristic peaks at 332 nm and 425 nm were used to characterize E. coli, respectively, and the sterilization rate of silver colloid was calculated. Sterilization rate (%) =(Ie-I)/Ie, where Ie is the E. coli fluorescence intensity, and I is the E.coli fluorescence intensity after addition of Nanometal, which was shown in figure 2.

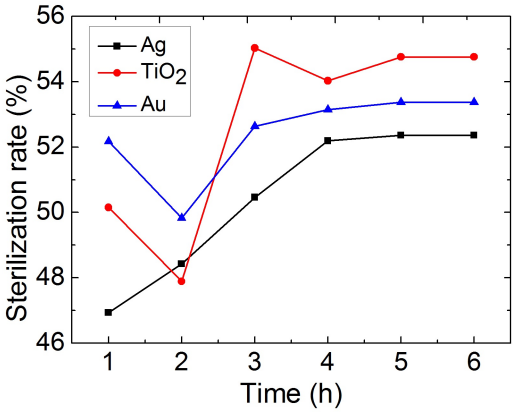

Fig 2. Sterilization rate with different time

It can be seen from figure 2 that when the same amount of nano-solution is added, the three nano-metals do not have the same sterilization time. The silver nanoparticle solution and E. coli reaction after 4h to complete the sterilization effect. The titanium dioxide solution and E. coli reaction after 3h to complete the sterilization effect. The gold Nanoparticle Solution and E. coli reaction after 3h to complete the sterilization effect. This shows that when the three kinds of nanometals have same amount and environment, their sterilization rate is Ag <TiO2 = Au.

2) Effects of addition amount and temperature on the sterilization

Three kinds of nanometals were added to E. coli with a fixed reaction time of 4 h. Measuring thesterilization effect with different amount and temperature. As shown in Figure 3.

As can be seen in Figure 3, the amount of nano metal added is proportional to the sterilization effect, but the temperature is not directly proportional to the sterilization effect. Silver Nanoparticle sterilization efficiency is easily affected by temperature, and Silver Nanoparticle sterilization have better sterilization effect with the higher the temperature. Titanium dioxide sterilization efficiency is less affected by temperature, and Titanium dioxide sterilization have lower sterilization effect with the higher the temperature. Gold Nanoparticle Solution is affected scarcelyby temperature.

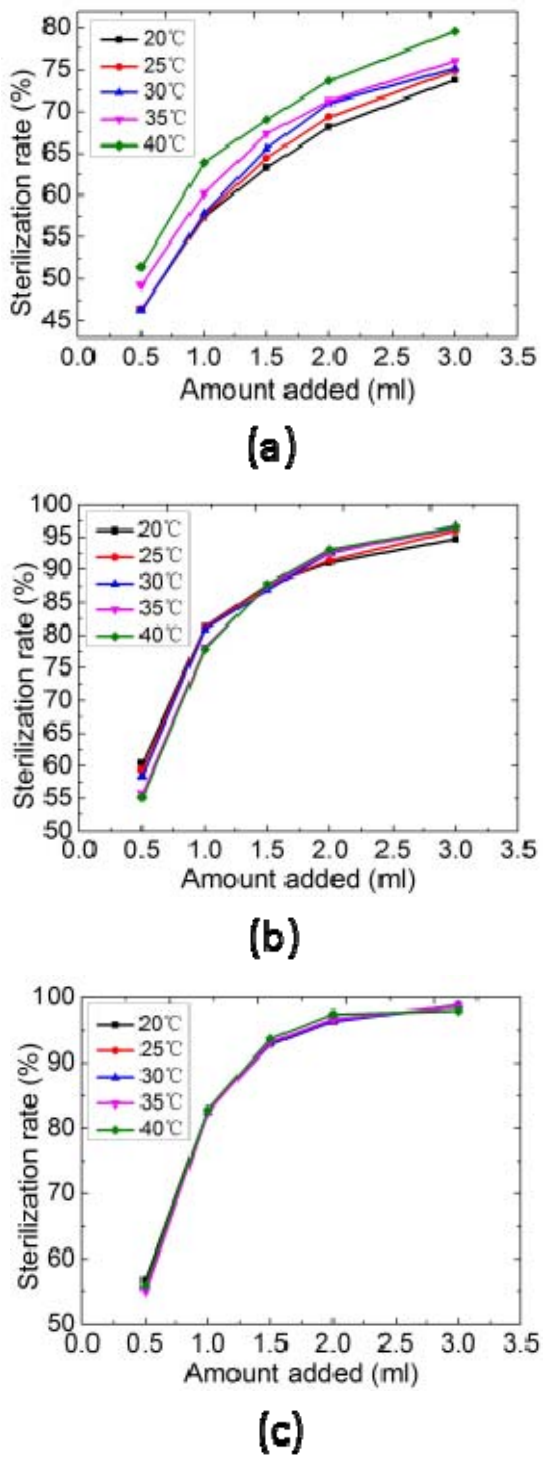

Fig 3. Effect of addition amount and temperature on sterilization effect

(a) Silver nanoparticles, (b) Titanium dioxide, (c) Gold nanoparticles

Among the three kinds of nanometals, silver nanoparticles have the worst sterilization effect and it are affected easily by temperature. The sterilization process is basically completed in 4 hours, and the sterilization effect is proportional to the amount of addition. Titanium dioxide has better sterilization effect and it is not affected easily by temperature. The sterilization process is basically completed in 3 hours, and the sterilization effect is proportional to the amount of addition. Gold nanoparticles have excellent sterilizing effect, and it can not be affected by temperature. The sterilization process is basically completed in 3 hours, and the sterilization effect is proportional to the amount of addition.

IV. Conclusion

In this work, we study the luminescent properties of E. coli and the sterilization of three nanometals. We find that the two emission peaks of E. coli are located at 332 nm and 425 nm with excitation light near 289nm. The emission peak at 332nm is the result of the superposition of tyrosine and tryptophan of amino acids in the protein, and the emission peak near 425nm is generated by the superposition of nucleic acid and NADH. The effect of the three nanometals on the killing of E. coli showed that the gold nanoparticles have excellent sterilizing capabilities and it is more suitable for sterilization than silver nanoparticles and titanium dioxide. So, nanogold is more suitable for sterilization.

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