Photothermal effect of Copper Sulfide Nanoparticles in Skin Defects in mice

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Abstract. Photothermal therapy is a non-invasive or minimally invasive, controllable, and has with good antimicrobial efficiency, while the main thing about skin defects is the prevention of bacterial infection as well as rapid repair and healing. Compared with drug therapy, photothermal therapy has the advantages of convenience and no side effects. In this paper, we mainly synthesize a copper sulfide nanoparticle with photothermal effect and wrap it in biocompatible mesoporous silica to make it work to the maximum extent. We It can be concluded from the data obtained from the defect experiments in mice animals that the skin defects in the mice after irradiated with NIR light take took less time to repair and heal than those mice not irradiated with NIR, so it can be shown that Photothermal photothermal treatment plays an important role here.

1. Introduction
The skin is the largest organ of the body and has the function of preventing microbial infections [1-3], maintaining fluid and electrolyte homeostasis, and resisting physical and chemical damage. When the integrity of the skin is disrupted, such as burns, skin defects, chronic ulcers, etc., skin trauma is formed, which can lead to microbial invasion, disruption of the internal environment and damage to the health of the body. Thus, skin defects should not be underestimated. Skin defects are always in need of medication to keep them free from bacterial infection and to accelerate the growth of skin in the defective areas. Therefore, it is important to find a treatment method that has no side effects and quick results.

Photothermal therapy [4,5] has become a hot research topic in recent years, which has many points such as non-invasive or minimally invasive, controllable, good antibacterial efficiency, no drug resistance and target control. The mechanism of photothermal therapy is that the photothermal agent can absorb a large amount of light energy and release it in the form of heat under the irradiation of near-infrared laser, and then act on the bacteria to kill them, thus accelerating the healing of the skin defect.

The main material used for photothermal therapy in this paper is Copper Sulfide Nanoparticle (CuS NPs) [6-8]. This is because copper sulfide is a good photothermal material when used in the treatment of bacterial infections. Copper sulfide has excellent photostability and there is no significant decrease in its light absorption over a longer period of time. Another great advantage of copper sulfide is its low cost, so it is a good principle, but CuS NPs have the disadvantage of hydrophobicity, so we encapsulated CuS NPs into mesoporous silica. Mesoporous silica [9] has good biocompatibility and it can be a good carrier for CuS NPs.
2. Experimentation

2.1. Synthesis of CuS NPs
Dissolve 5 g of polyethylene glycol and 5 mmol of CuCl₂·2H₂O in 50 mL of deionized water, stir thoroughly until clear and transparent, and then dissolve 50 mmol of sodium sulfide nonahydrate in 200 mL of deionized water to obtain an aqueous solution of sodium sulfide. Add 20 ml of sodium sulfide solution to the above copper chloride solution and stir to make it fully react. Then it was put into the oven and kept at 50°C for four hours, and finally it was washed by deionized water and centrifuged three times, and dried in the oven at 45°C to obtain CuS NPs [10].

2.2. Synthesis of CuS@SiO₂
0.2 g of CuS NPs were dissolved in 20 ml of deionized water, 50 ml of ethanol and 15 ml of deionized water were mixed and added to the CuS NPs solution, then 20 ml of ammonia and 0.2 g of cetyltrimethylammonium bromide were added and stirred under a magnetic stirrer for half an hour, after which 5 ml of Tetraethyl orthosilicate (TEOS) was added and stirring was continued for half an hour, followed by stirring at 50°C for 4 hours. Wash with ethanol and deionized water, and dry at 60°C to obtain CuS@SiO₂.

2.3. Near-infrared photothermal testing
CuS NPs and CuS@SiO₂ were dissolved in PBS and the solutions were placed in a centrifuge tube, which was irradiated with NIR light and used for in vitro photothermal testing; the solutions were injected subcutaneously into mice and irradiated with NIR light and used for in vivo testing.

2.4. Photothermal treatment of skin defects
The mice skin defect repair experiment was divided into control group and experimental group, the defect area of control group was coated with PBS buffer solution, and the surface of experimental group was coated with CuS@SiO₂ solution, then the two groups were treated with photothermal irradiation to observe the changes after 7 days.

3. Results and Discussion

3.1. Characterisation of CuS NPs and CuS@SiO₂

![Figure 1](image1.png)

Figure 1 (a) Transmission electron micrograph of CuS NPs; (b) Transmission electron micrograph of CuS@SiO₂
These two figures are transmission electron microscopy images of CuS NPs and CuS@SiO$_2$, respectively. From (a), it can be seen that the size of CuS NPs is around 3 nm, and the small size of nanoparticles with photothermal effect makes the photothermal effect better, so this synthesis is successful. From (b), we can see the obvious encapsulation, which can indicate that the mesoporous silica wrapping CuS NPs is successful and the size is around 300 nm, which is also well in line with the requirement of the material size.

3.2. BET characterization of CuS@SiO$_2$

| parameter                                      | Value     |
|------------------------------------------------|-----------|
| BET surface area (m$^2$/g)                     | 245.0141 m$^2$/g |
| average pore diameter (nm)                     | 5.45968 nm |
| Single point adsorption total pore volume of pores (cm$^3$/g) | 0.338292 cm$^3$/g |

Figure 2. N$_2$ absorption–desorption isotherms

The pore size of CuS@SiO$_2$ measured using the nitrogen adsorption specific surface area method is 5.45968 nm and the specific surface area is 245.0141 m$^2$/g. This indicates that the pore size of mesoporous silica is relatively ideal, so that drug loading and targeting studies can be considered later.

3.3. The photothermal properties of CuS@SiO$_2$

Figure 3. Temperature rising curves of CuS@SiO$_2$ in vitro and in vivo
To study the photothermal effect of CuS@SiO$_2$, we conducted in vivo and in vitro irradiation of near-infrared light with 808 nm at a power of 1.5 W/cm$^2$. From the figure, we can see that the heating rate of CuS@SiO$_2$ is fast, from 35.5°C to 57.6°C in less than two minutes in vitro, and the same experiment in vivo time, warming from 35.5°C to 45.9°C. From these data, it can be concluded that CuS@SiO$_2$ has a good photothermal effect.

**Figure 4.** (a) In vitro NIR irradiation experiment; (b) In vivo NIR irradiation experiment

3.4. *Experimental analysis of skin defect repair*

**Figure 5.** (a) (b) shows the initial skin defect picture; (c) (d) shows the skin defect picture after seven days
In the skin defect experiment, (a) is the experimental control group, only PBS solution was added to the skin surface for NIR light irradiation, and (b) is the experimental group, CuS@SiO$_2$ solution was applied to the skin defect surface for NIR light irradiation. (c) (d) shows the recovery of the experimental control group and the experimental group after seven days, respectively. From the figure, we can clearly see that the area of the repaired skin defect in the experimental group is larger than that in the control group, and we can clearly see that the skin defect area in the experimental group starts to crust. Therefore, it can be shown that the photothermal treatment effect of CuS@SiO$_2$ with NIR irradiation is still good.

4. Conclusion
In this paper, we synthesized CuS NPs and used the excellent biocompatibility of mesoporous silica as a carrier for CuS NPs, so that it can be used for skin defect repair without considering whether it has side effects. In this paper, we concluded that the photothermal treatment of CuS@SiO$_2$ is effective, and the pore size of the mesoporous silica we prepared is also ideal, so it can be used as a drug carrier later, which can achieve the effect of photothermal-drug dual treatment.

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