Supporting Information

One-pot synthesis of PEGylated plasmonic WO_{3-x}@Eugenol nanoflowers with NIR-controllable antioxidant activities for synergetic combating bacteria biofilm infection

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1. **Experimental Section**

1.1 Apparatus and characterization

The morphology of the nanomaterials was characterized by transmission electron microscope (TEM, JEOLJEM-1230), Scanning electron microscopy (SEM) and energy-dispersive spectroscopy (EDS) analysis were measured by S-4800 FE-SEM (Hitachi, Japan). The crystalline structure was determined by a Bruker D8 X-ray diffraction (XRD) system. The chemical composition was measured by X-ray photoelectron spectrometer (XPS), employing an Axis Ultra DLD X-ray photoelectron spectrometer equipped with Al Kα X-ray as the excitation source (1486.6 eV). Fourier transform infrared (FTIR) spectra was recorded on the wavenumber range of 400-4000 cm^{-1} with a Vetex70 (Bruker Corp, Germany) instrument using the KBr pellet method. UV-visible absorption spectra were obtained using a UV-2550 UV-vis spectrophotometer (Shimadzu, Japan).

1.2 SEM characterization of bacteria

The bacterial morphology was characterized by SEM. After antibacterial experiments, the bacteria were collected by centrifugation and fixed overnight with 2.5% glutaraldehyde, followed by dehydrated sequentially in ethanol solutions with different concentrations (30%, 50%, 70%, 90%, and 100%, v/v) for 10 min. Finally, the samples were freeze-dried with a critical point dryer, sputter-coated, and observed by SEM.

1.3 Biofilm formation and crystal violet assay

First, 10 μL of bacterial suspension (10^8 CFU mL^{-1}) and 90 μL tryptophan soybean broth (TSB) was added to sterile 96 well polystyrene microplate and incubated at 37 ℃ for 48 h to obtain mature biofilm. Then, crystal violet experiment was used to evaluate the ability of the material to inhibit biofilm. The obtained biofilm was washed with sterile normal saline, and then incubated at 37 ℃ for 24 h with different treatment methods (without / with material and without / with light).

After that, wash with sterile normal saline and dry in the air. Then, add 100 μL crystal violet to each well (0.1%, W/ V, sterile water as solvent), dye for 5 min, wash the excess dye with sterile water, dissolve the dyed biofilm in the hole in acetic acid solution (33% v/V), and finally detect the absorbance at 595 nm with enzyme labeling instrument, which represents the quality of biofilm.

1.4 Evaluation of biofilm elimination by MTT method

Briefly, The obtained biofilm was washed with sterile normal saline, then the material was added for light or no light treatment, and incubated at 37 ℃ for 24 hours. After washing with physiological saline, add 100 μL of MTT (5 mg mL^{-1}) solution to each well and incubate at 37 °C for 3 h. Add another 100 μL dimethyl sulfoxide (DMSO) dissolves the blue purple precipitate. Finally, the absorbance of each well was measured by microplate reader at 592 nm.

1.5 Confocal Laser Scanning Microscopy (CLSM) Assessment of Biofilm Disruption

Briefly, The biofilm was treated with different treatments, the three was washed to remove non-adherent bacteria, and then incubated with fluorescein diacetate (FDA). Finally, the biofilm was imaged in 3D by confocal laser scanning microscopy (CLSM) (Nikon A1R+, Japan).

1.6 Cell toxicity assay

The MTT method was used to detect the in vitro cytotoxicity of different concentrations of PEG-WO_{3-x}@EL NFs (0, 10, 50, 75 and 100 μg/mL) on NIH 3T3 cells. Place the mouse embryonic fibroblasts (NIH 3T3) frozen in a liquid nitrogen tank in a 3T3 cell culture medium and culture and activate them at 37 °C and 5% CO_2. After the cells have grown to the logarithmic phase, treat the cells with trypsin-EDTA digestion solution, pipette the cells repeatedly with 3T3 cell culture medium to prepare a cell suspension, and inoculate the cells into 96-well cells according to 2×10^5 cells per well. In the culture plate, incubate at 37 °C for 24 h. Then the cells were dealed nanomaterials for 24 h. After treatment, the cells were washed three times with

PBS, and 10 μL of MTT solution (5 mg/mL) was inject into each well. After incubating at 37 °C for 4 h, remove the supernatant liquid, add 100 μL of DMSO to each well. The absorbance was determined at 490 nm with microplate.

2. Results

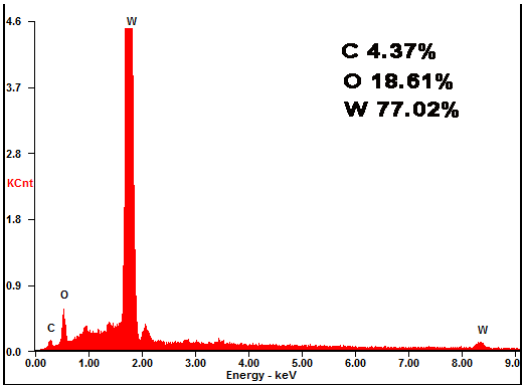

Fig. S1. Energy spectra of PEG-WO_{3-x}@EL NFs.

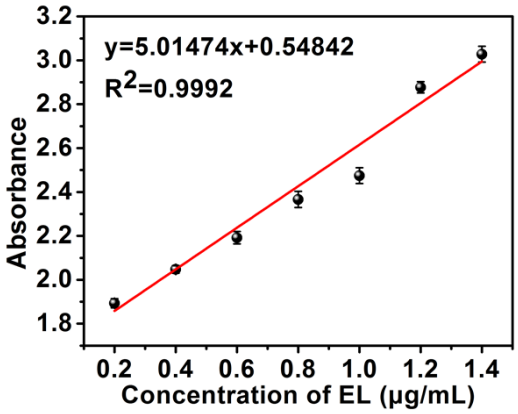

Fig. S2. Standard curve of absorbance versus EL concentration.

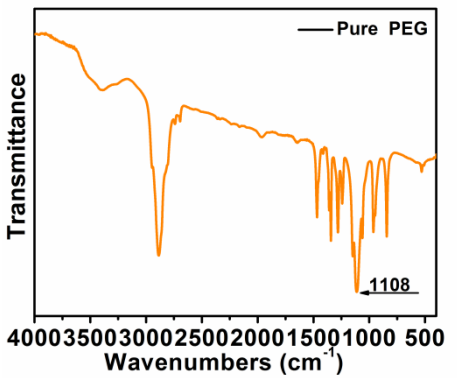

Fig. S3. FI-IR spectra of pure PEG.

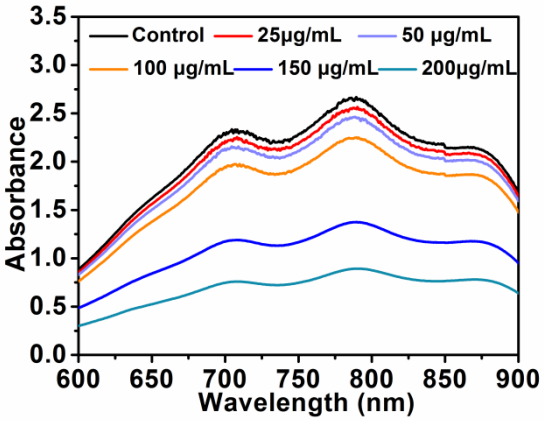

Fig. S4. ABTS measured the absorption curves of the material in different concentrations.

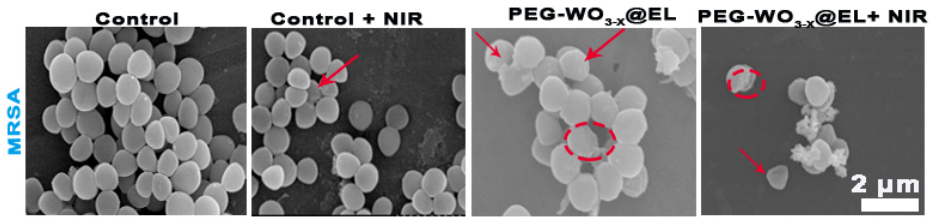

Fig. S5. SEM images of *MRSA*. Bacterial deformation or destruction at red arrows.

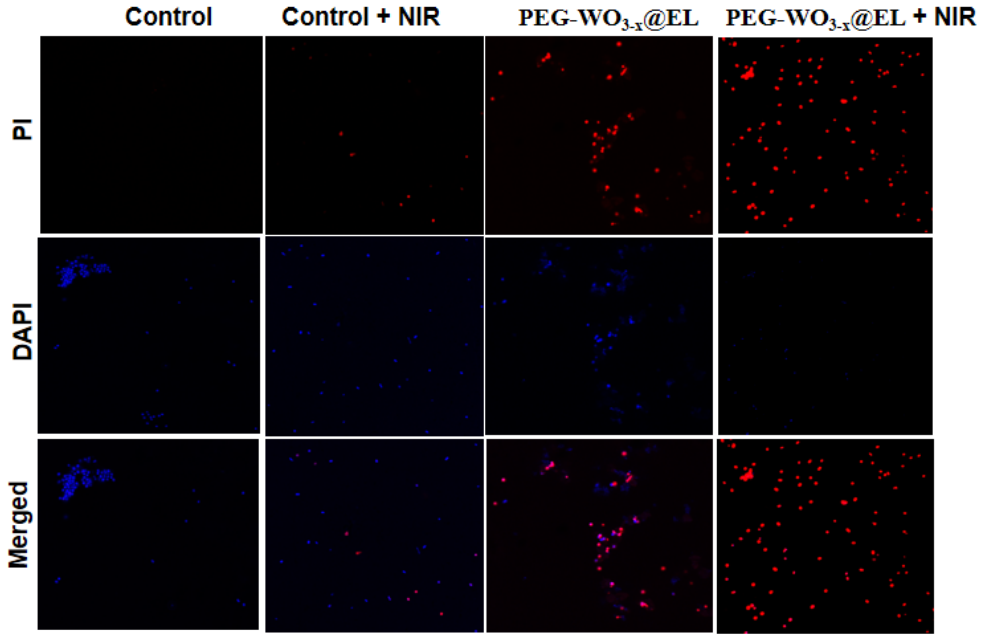

Fig. S6. Fluorescence image of *MRSA*.

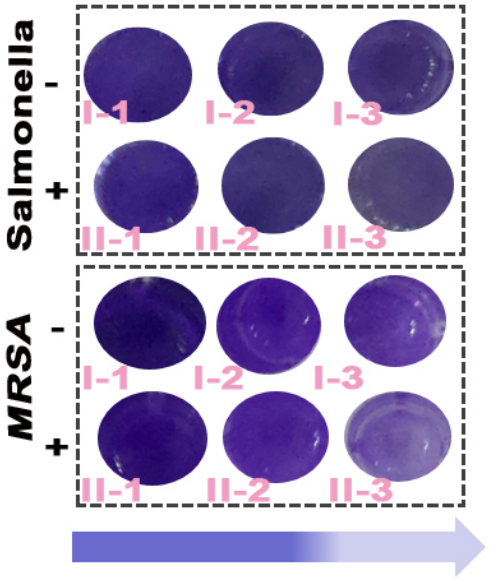

Fig. S7. Pictures of *Salmonella* and *MRSA* biofilms stained with crystal violet under different treatments. (I-1 - I-3 were control groups, PEG-WO_{3-X} NFs group, and PEG-WO_{3-x}@EL NFs group respectively, and II - 1-II-3 were control + NIR groups, PEG-WO_{3-X} NFs + NIR group, and PEG-WO_{3-x}@EL NFs + NIR group respectively).

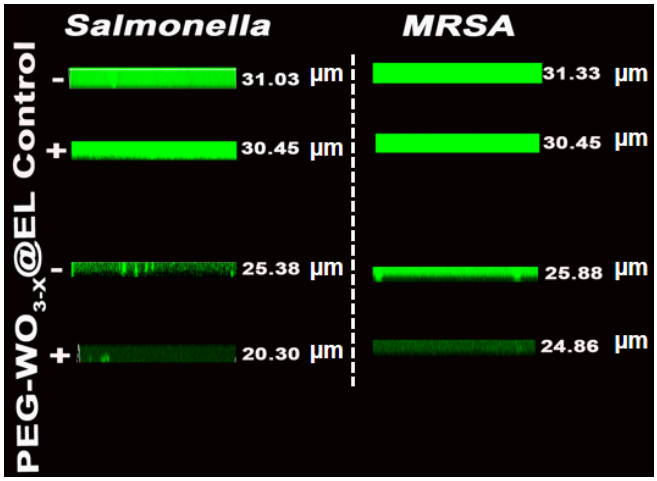

Fig. S8. Confocal laser scanning microscope (CLSM) showed that the material inhibited the thickness of biofilm under different treatments.