

Supplemental Information

Electrospun flexible magnesium-doped silica bioactive glass nanofiber membranes with anti-inflammatory and pro-angiogenic effects for infected wounds

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Methods

1. Tensile test of electrospun SiO₂/MgO membranes

Electrospun SiO₂/MgO membranes were tailored into strip specimens (10 × 40 mm) for uniaxial tensile tests. Ends of strip specimens were mounted in the grips of a universal testing machine (Instron 5567, Norwood, MA) with a 200 N load cell. Uniaxial tensile testing of samples was performed at a crosshead speed of 10 mm/min until failure. Ultimate tensile strength (UTS) was determined by the maximum tensile strength before failure and Young's modulus was calculated as the slope of the initial 3% linear portion from the stress-strain curve (n = 4 for each group).

2. Preparation of extract from electrospun SiO₂/MgO membrane

Immerse electrospun SiO₂/MgO-2% membrane in 20 mg/mL medium (DMEM) without FBS and incubate at 37 °C for 24 h. Filter through a 0.22 μm filter, collect the sterile supernatant, add LPS and IFN-γ, and prepare a membrane incubation medium containing 1000 μg/mL LPS and 20 ng/mL IFN-γ.

3. Quantitative real-time polymerase chain reaction (qRT-PCR)

RNA was extracted from wound tissue for qRT-PCR analysis. Total RNA was isolated from wound tissue using Trizol (Sangon Biotech Co., Ltd., Shanghai, China). cDNA was synthesized using the RevertAid First Strand cDNA Synthesis Kit (Thermo Fisher Scientific). Quantitative RT-PCR was performed on an Applied Biosystems™ 7500 Real-Time PCR System using NovoStart® SYBR qPCR SuperMix Plus (Novoprotein Scientific Inc., Shanghai,

China) and analyzed by the comparative Ct quantification method ($\Delta\Delta C_t$). The sequences of primers are shown in Table S1. The expression level of the gene was correlated with glyceraldehyde 3-phosphate dehydrogenase (GAPDH) and normalized to the level of the control group (uninfected group).

4. Histological analyses

Histological analysis was performed on hematoxylin and eosin (H&E) and Masson's Trichrome staining, the standard protocol for these histological stains was first to work with tissue sections according to the manufacturer's instructions and to acquire the tissue with a slide scanner (PRECICE 500X) study images.

5. Immunostaining

The tissue sections were successively dewaxed with xylene, dehydrated with ethanol, and rehydrated with deionized water. After antigen retrieval, tissue sections were first blocked with 5% bovine serum albumin solution to remove paraffin sections. Tissue sections were then incubated with primary antibodies, washed with phosphate-buffered saline, and then incubated with the corresponding secondary antibodies.

Table S1. Primer sequences of genes of mouse tissue for qRT-PCR analysis.

Gene name	Primer sequence (5'-3')
COL-1	F: 5'-GCTCCTCTTAGGGGCACT-3' R: 5'-CCACGTCTCACCATTGGGG-3'
MMP-1	F: 5'-GAGTCCATGTTCATGGTGGGA-3' R: 5'-TTGGGAAGAGCTCCTCTGTT-3'
TGF- β 1	F: 5'-CTTCAATACGTCAGACATTCGGG-3' R: 5'-GTAACGCCAGGAATTGTTGCTA-3'
VEGF	F: 5'-GCACATAGAGAGAATGAGCTTCC-3' R: 5'-CTCCGCTCTGAACAAGGCT-3'

TNF- α	F: 5'-CCTGTAGCCCACGTCGTAG-3' R: 5'-GGGAGTAGACAAGGTACAACCC-3'
IL-1 β	F: 5'-TGCCACCTTTTGACAGTGATG-3' R: 5'-AAGGTCCACGGGAAAGACAC-3'
IL-10	F: 5'-GCTCTTACTGACTGGCATGAG-3' R: 5'-CGCAGCTCTAGGAGCATGTG-3'
Arg-1	F: 5'-CTCCAAGCCAAAGTCCTTAGAG-3' R: 5'-AGGAGCTGTCATTAGGGACATC-3'
GAPDH	F: 5'-GTGACGTTGACATCCGTAAAGA-3' R: 5'-GCCGGACTCATCGTACTCC-3'

Results

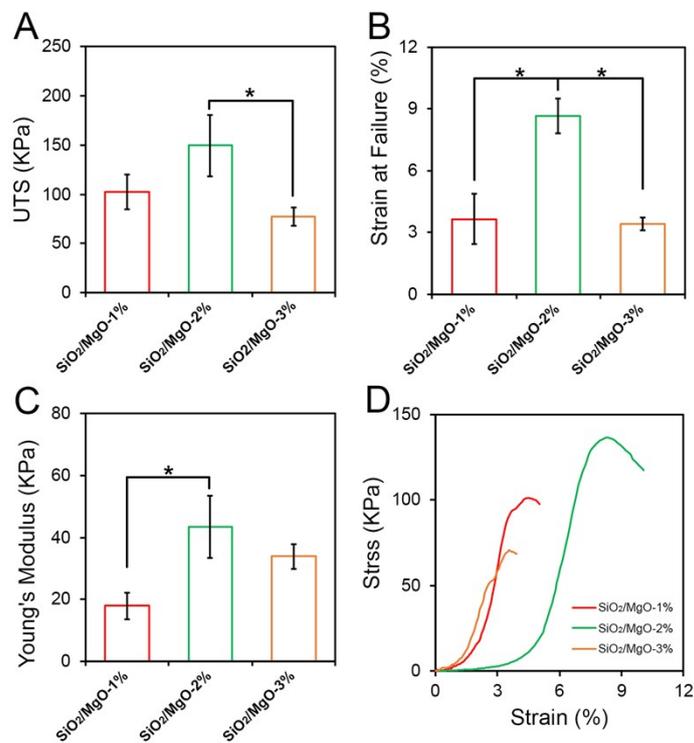


Fig. S1 The SiO₂/MgO-2% nanofiber membrane has greater UTS (A), breaking strains (B) and Young's moduli (C), and the stress-strain curve (D) than the other SiO₂/MgO membranes. One-way ANOVA with Tukey's post hoc test, n = 4, * indicates $p < 0.05$.