Supporting Information

Toxicity and mechanism of mesoporous silica nanoparticles to the eyes

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Fig. S1. Characterization of FITC-MSiNPs. A) Nitrogen adsorption-desorption isotherm of the as-prepared monodispersed FITC-MSiNPs. B) Thermogravimetric analysis pore volume of FITC-MSiNPs.



Fig. S2. The adsorption of metal ions (Ag⁺) loaded on MSiNPs was determined by UVvis spectrophotometry. A) The calibration curve of Ag⁺. B) The evolution of UV-vis spectra of Ag⁺ adsorption by MSiNPs at different concentrations.



Fig. S3. The culture and identification of hCECs. A-C) Primary culture of human cornea explant. hCECs expanded from the human cornea explant which was indicated by asterisk. Scale bar, 50 μ m. D-F) Immunostaining of CK12 (red) in P₂ hCECs. Nuclei were stained by DAPI (blue). Scale bar, 50 μ m. G-I) Primary culture of Immunostaining of CK15 (red) in P₂ hCECs. Nuclei were stained by DAPI (blue). Scale bar, 50 μ m.



Fig. S4. The cytotoxicity of hCECs exposure to the free Ag⁺ for 24 h. Bottom line indicates the corresponding MSiNPs-Ag⁺ concentration to free Ag⁺.



Fig. S5. The cellular uptake of Ag^+ in MSiNPs- Ag^+ group. Column chart illustrating the mean Ag^+ amount (ng/10⁴ cell) obtained by using ICP-MS from washed hCECs after exposed to MSiNPs- Ag^+ .



Fig. S6. The cell apoptosis of hCECs exposure to MSiNPs, MSiNPs-Ag⁺. A) Proportion (%) of hCECs exposed to MSiNPs and MSiNPs-Ag⁺ that indicate live cells (Annexin V + PI). B) Proportion (%) of hCECs exposed to MSiNPs and MSiNPs-Ag⁺ that show early or late apoptosis. n=3 in each group. Data are the mean ± SEM from three independent experiments. **P < 0.01, ***P < 0.001 using one-way ANOVA and post-hoc Tukey's test.



Fig. S7.Distribution of DEGs and KEGG pathway analysis of hCECs exposed to MSiNPs and MSiNPs-Ag⁺. A) Pie chart of all DEGs in the MSiNPs group compared with the control group, blue: up-regulated genes in the MSiNPs group; red: down-regulated genes in the MSiNPs group when the hCECs were exposed. B) Same with A) but for hCECs exposed to MSiNPs-Ag⁺. C) The differentially expressed mRNAs in the control group and MSiNPs group were classified and clustered. Each row represents a single mRNA and each column represents one tissue sample. Red, high relative expression; blue, low relative expression. D) The differentially expressed mRNAs in the control group and MSiNPs-Ag⁺ group were classified and clustered. P-values and Q-values (MA-plot) represent the significance of the differences (fold change \geq 2 and adjusted P-value \leq 0.05).



Fig. S8. The 15 most significantly altered pathways in the Kyoto Encyclopedia of Genes and Genomes (KEGG). A) The 10 most significantly changed biological processes in the KEGG enrichment analyses of MSiNPs group compared with control. B) The 10 most significantly changed biological processes in the KEGG enrichment analyses of MSiNPs-Ag⁺ group compared with control. Q-values and P-values represent the level of significance of enrichment.



Fig. S9. Evaluation of adsorption capacity of MSiNPs and MSiNPs-Ag⁺. Percentage changes of adsorbed BSA to total BSA over time in MSiNPs group (the red curve, 2 mg/mL) and MSiNPs-Ag⁺ group (the black curve, 2 mg/mL).