Supplementary Information

Synthesis of Nanostructured Silica/Hydroxyapatite Hybrid Particles Containing Amphiphilic Triblock Copolymer for Effectively Controlling Hydration Layer Structures with Cytocompatibility

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Scheme S1. Illustration of the resistive pulse curve to indicate the maximum resistance change ($\Delta R$) from the baseline level with the applied voltage time. Inset: illustration of the cross-section of membrane pore structure with the passing of particle along.
Figure S1

Figure S1. (a) UV-Visible absorption spectra of the Ab in ultrapure water with the different concentrations, and (b) their calibration curves between the concentration and absorbance ($R^2$=0.99913).
Figure S2. FT-IR spectral deconvolution results of the amide I absorption band of Ab, and their secondary structural components ((i): β-sheet, (ii): random, (iii): α-helix, (iv): turn, (v)(vi): β-turn), and their (vii) re-synthesized and (viii) raw spectra.
Figure S3. Possible illustration of folding protein structures at the (a) native state and (b) denatured states.
**Table S1**

Resultant chemical compositions of HA and the nanostructured MS/HA hybrid particles, and their Ca/P and Ca/Si molar ratios, which were measured by an XRF analysis.

<table>
<thead>
<tr>
<th></th>
<th>Si (mol %)</th>
<th>O (mol %)</th>
<th>Ca (mol %)</th>
<th>P (mol %)</th>
<th>The others (mol %)</th>
<th>Ca/P (molar ratio)</th>
<th>Ca/Si (molar ratio)</th>
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<tbody>
<tr>
<td>HA</td>
<td>–</td>
<td>52.8</td>
<td>17.9</td>
<td>10.1</td>
<td>9.4</td>
<td>1.77</td>
<td>–</td>
</tr>
<tr>
<td>MS/HA-0</td>
<td>12.3</td>
<td>58.2</td>
<td>9.5</td>
<td>5.1</td>
<td>14.8</td>
<td>1.86</td>
<td>0.77</td>
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<tr>
<td>MS/HA-1</td>
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<td>58.7</td>
<td>9.5</td>
<td>5.1</td>
<td>14.0</td>
<td>1.87</td>
<td>0.77</td>
</tr>
<tr>
<td>MS/HA-2</td>
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<td>61.5</td>
<td>9.6</td>
<td>5.1</td>
<td>10.8</td>
<td>1.87</td>
<td>0.82</td>
</tr>
<tr>
<td>MS/HA-3</td>
<td>12.1</td>
<td>58.9</td>
<td>10.0</td>
<td>5.3</td>
<td>12.0</td>
<td>1.87</td>
<td>0.83</td>
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</table>
Figure S4. (a) solid state $^{29}$Si–NMR spectral deconvolution results of MS and the nanostructured MS/HA hybrid particles, indicating the separated spectra of (1) $Q_2$ (i.e., two Si–O–Si and two Si–OH bonds) at –93±2 ppm, (2) $Q_3$ (i.e., three Si–O–Si and one Si–OH bonds) at –102±2 ppm and (3) $Q_4$ (i.e., four Si–O–Si bonds) at –111±2 ppm, and (4) their re-synthesized and (5) raw spectra. (b) $(Q_2+Q_3)/Q_4$ ratios of the nanostructured MS/HA hybrid particles with the concentration of P123. Here, the $(Q_2+Q_3)/Q_4$ ratio of MS is 0.98.
Scheme S2

Illustration of the possible interfacial bonding formation between the MS and HA phases.
Figure S5. XRD pattern of the MS at the lower 2θ region.
**Scheme S3**

Illustration of the possible interfacial bonding reaction between the MS and HA phases.

\[
\text{TEOS} \quad \text{SiO}_4^{4-} \quad \text{HA} \quad \text{Si} \quad \text{O} \quad \text{OH} \\
\text{O–H dissociates} \quad (pK_a=6.5 \sim 9.8) \\
\text{HPO}_4^{2-} \quad (pK_a=12.38) \\
\text{Silicate ion partially replaced} \\
\text{Ca}_{10} (\text{PO}_4)_6 (\text{OH})_2 + x\text{SiO}_4^{4-} \to \text{Ca}_{10} (\text{PO}_4)_{6-x} (\text{SiO}_4)_x (\text{OH})_{2-x} + x\text{PO}_4^{3-} + x\text{OH}^{-}
\]
Figure S6. FT-IR spectral deconvolution results of three hydration layers formed on the particles. The separated (I), (II) and (III) peaks are attributed to free, intermediate and nonfreezing water molecular states, respectively, and (IV) their re-synthesized and (V) raw spectra.
**Figure S7**

FT-IR spectral deconvolution results of six water-interactive states formed on the particles, indicating the separated spectra of (1) asymmetric O–H stretching vibration between waters, (2) deformation O–H vibration of water, stretching O–H vibration of water on Si–OH, (3) symmetric O–H stretching vibration of water, (4) non-stretching O–H vibration of water on Si–OH, (5) hydrogen bonded stretching O–H vibration, stretching O–H vibration of water on Si–OH and (6) non-stretching O–H vibration of structural OH in HA, respectively, and (7) their re-synthesized and (8) raw spectra.
Figure S8

Figure S8. Adsorbed Ab amount (a) per weight and (b) per BET surface area on the particles ($n$=3, mean ± S.D.).
**Figure S9**

FT-IR spectral deconvolution results of the amide I bands of the adsorbed Ab on the particles to provide the secondary structural components (i): β-sheet, (ii): random, (iii): α-helix, (iv): turn, (v)(vi): β-turn), and their (vii) re-synthesized and (viii) raw spectra.
Figure S10. The component ratios on the secondary structures of the amide I bands in the adsorbed Ab on the particles, and (inset) their averaged ratios of the reference samples between HA and MS and the hybrid particles between MS/HA-0, MS/HA-1, MS/HA-2 and MS/HA-3. ** indicates the students’ t-test values as $P < 0.01$ based on the significant difference of (iii) α-helix component between the reference samples and the hybrid particles ($n=3$, mean ± S.D.).
Figure S11. (a) Adhered osteoblast-like cell density changes on TCPS dish without particle addition and with adding APACERAM®, HA, MS/HA-0 and MS/HA-3 (n=10, mean ± S.D.), and (b) the representative optical microscope images of the cells at the culture time of 72 h, and the added particles were observed around the cells.