Supporting Information

Hierarchically stimuli-responsive nanovectors for improved tumor penetration and programmed tumor therapy

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Scheme S1. Synthetic procedures of (A) cisplatin prodrug, c, c, t-\([\text{Pt}(\text{NH}_3)_2\text{Cl}_2(\text{OH})(\text{O}_2\text{CCH}_2\text{CH}_2\text{CO}_2\text{H})]\), and (B) PAMAM-Pt(IV) or PAMAM-FITC dendrimers.
Figure S1. FTIR spectra of cisplatin (a), oxoplatin (b), c, c, t-[Pt(NH$_3$)$_2$Cl$_2$(OH)(O$_2$CCH$_2$CH$_2$CO$_2$H)] (c) and PAMAM-Pt(IV) (d), respectively.

The characteristic absorption peaks at 3283 cm$^{-1}$, 1302 cm$^{-1}$ and 805 cm$^{-1}$ were ascribed to the stretching vibration of N-H bonds from cisplatin. The absorption peak at 513 cm$^{-1}$ was characteristics of V$_{pt-N}$, which corresponds to the cis-structure of platinum. After oxidation with hydrogen peroxide, c,c,t-[Pt(NH$_3$)$_2$Cl$_2$(OH)$_2$] showed a sharp, intense O–H stretching band at 3516 cm$^{-1}$. Meanwhile, additional absorption peaks at 1040 cm$^{-1}$ and 558 cm$^{-1}$ were ascribed to the Pt-OH bend and Pt–N stretch vibrations, respectively.$^{51}$ The observations imply that cisplatin was successfully
oxidized into oxoplatin. The $\text{c,c,t-[Pt(NH}_3\text{)}_2\text{Cl}_2(\text{OH})(\text{O}_2\text{CCH}_2\text{CH}_2\text{CO}_2\text{H})]}$ showed additional strong absorption peak at 1709 cm$^{-1}$, which was assigned to the carbonyl (C=O) in carboxyl groups.$^{52}$ New peaks at 1253 cm$^{-1}$ and 1118 cm$^{-1}$ were attributed to stretching vibration of the C-O in ester bonds. Meanwhile, characteristic peak corresponding to the $-\text{CH}_2-$ at 2913 cm$^{-1}$ was also observed, indicating the successful preparation of $\text{c,c,t-[Pt(NH}_3\text{)}_2\text{Cl}_2(\text{OH})(\text{O}_2\text{CCH}_2\text{CH}_2\text{CO}_2\text{H})]}$. The final PAMAM-Pt(IV) prodrug was prepared via amide reaction of between PAMAM and $\text{c,c,t-[Pt(NH}_3\text{)}_2\text{Cl}_2(\text{OH})(\text{O}_2\text{CCH}_2\text{CH}_2\text{CO}_2\text{H})]}$. The IR spectrum of PAMAM-Pt(IV) displayed a broad absorption peak of N–H bonds at 3289 cm$^{-1}$. The peaks at 2829, 2940, and 3084 cm$^{-1}$ corresponding to the stretching vibrations of the C–H bonds in PAMAM unit are also observed. The peak at 1434 cm$^{-1}$ was contributed to the bending vibrations of the C–H bonds in dendrimer. Moreover, new peaks at 1645 and 1554 cm$^{-1}$ were attributed to stretching vibration of amide I and amide II of PAMAM dendrimer structure. All results indicate that PAMAM-Pt(IV) prodrug was successfully synthesized.$^{54}$
Figure S2. Relationship between pH and zeta potential: changes of zeta potentials of HMSN-CS(DMA)/PAMAM-Pt at different pH points.
**Figure S3.** Concentration-dependent cytotoxicity of drug-freed nanosystems.
Figure S4. Quantitative fluorescence intensity analysis after cells with HMSNs-CS(DMA)/ PAMAM-FITC or HMSNs-CS(SA)/PAMAM-FITC treatments at different pH.
Figure S5. Biodistributions of different platinum formulations (free cisplatin, PAMAM-Pt, HMSN-CS(SA)/PAMAM-Pt and HMSN-CS(DMA)/PAMAM-Pt) in the major organs of nude mice at 12 h (A) and 24 h (B). Data are presented as mean ± SD, n = 3.
Figure S6. Real-time measurements of average tumor-bearing mice weights after each treatment.
Figure S7. Histologic assessments of main organs of tumor bearing mice treated with saline (a), PAMAM-Pt (b), free GEM (c), HMSN@GEM-CS(SA)/PAMAM-Pt (d) and HMSN@GEM-CS(DMA)/PAMAM-Pt (e), respectively. The scale bar is 200 μm.
### Table S1. Survival of tumor-bearing mice after treatment with saline, PAMAM-Pt, GEM, HMSN@GEM-CS(SA)/PAMAM-Pt and HMSN@GEM-CS(DMA)/PAMAM-Pt.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>MST (days)</th>
<th>ILS (%)</th>
<th>Log-rank P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>saline</td>
<td>32.7</td>
<td>/</td>
<td>/</td>
</tr>
<tr>
<td>PAMAM-Pt</td>
<td>38.5</td>
<td>17.7</td>
<td>0.088</td>
</tr>
<tr>
<td>GEM</td>
<td>34</td>
<td>4</td>
<td>0.503</td>
</tr>
<tr>
<td>HMSN@GEM-CS(SA)/PAMAM-Pt</td>
<td>46.2</td>
<td>41.3</td>
<td>0.003</td>
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<tr>
<td>HMSN@GEM-CS(DMA)/PAMAM-Pt</td>
<td>56.7</td>
<td>73.4</td>
<td><strong>0.001</strong></td>
</tr>
</tbody>
</table>

Notes: n =6 for each treatment group; MST, mean survival time; ILS, increased life span; P2 value was calculated in comparison between HMSN@GEM-CS(DMA)/PAMAM-Pt with HMSN@GEM-CS(SA)/PAMAM-Pt.
References


