Supplementary Materials for

**Biological chemotaxis-guided self-thermophoretic nanoplatform augments colorectal cancer therapy through autonomous mucus penetration**

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Other Supplementary Material for this manuscript includes the following:

- Movies S1 to S3
**Supplementary Text**

**Materials:** Hexadecyltrimethylammonium bromide (CTAB), NaOH, tetraethyl orthosilicate (TEOS), 3-aminopropytrimethoxysilane (APTES), 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (EDC), N-Hydroxysuccinimide (NHS), dimethylformamide (DMF), and fluorescein isothiocyanate (FITC) were purchased from Aladdin Biochemical Technology Co., Ltd. (Shanghai, China). FITC labeled wheat germ agglutinin and PEG<sub>2000</sub>-SS-COOH were purchased from Ruixi Biotechnology Co., Ltd. (Xi’an, China). Mucin from porcine gastrointestinal tract were purchased from Ruixi Biotechnology Co., Ltd (Shanghai, China). Rabbit anti-protein A polyclonal antibody was purchased from Absin Bioscience Inc. (Shanghai, China). Anti-Ki67 mouse monoclonal antibody was purchased from Servicebio Technology Co., Ltd (Wuhan, China). SolarFast protein staining buffer and Cell Counting Kit-8 (CCK-8) was purchased from Solarbio Technology Co., Ltd. (Beijing, China). Goat Anti-rabbit IgG/HRP was purchased from Proteintech Group, Inc. (Chicago, USA). Mouse Tumor Necrosis Factor-α Enzyme-Linked ImmunoSorbent Assay Kit and Mouse Interleukin-6 Enzyme-Linked ImmunoSorbent Assay Kit were purchased from Beyotime Biotechnology Co., Ltd. (Shanghai, China).

**Instruments:** Platinum was deposited on the surface of mesoporous silica by vacuum evaporator (Leica EM ACE600, Germany). TEM images and EDS analysis were conducted by field emission transmission electron microscope (JEM-ARM300F, Japan). Dynamic light scattering and Zeta potential were measured using a Malvern Zetasizer Instrument. Nanoparticle movement and cell uptake were detected by confocal laser scanning microscope (CLSM, Leica SP-8, Germany).

**Cells and animals:** Caco-2 cells (human colon carcinoma Caco-2 cell line), HT29 cells (human colon carcinoma cell lines), and CT26 cells (murine colon carcinoma cell line) was obtained from the cell bank of Chinese Academy of Science (Shanghai, China). Mucus-producing HT29-CTX cells was obtained from laboratory storage. CT26 cell transfected with luciferase (CT26-luc) cells were kindly provided by Prof. Chao Fang (School of Medicine, Shanghai Jiaotong University, China). Caco-2, CT26 cells, and CT26-luc cells were cultured at RPMI 1640 medium (Solarbio, China) with 10% fetal bovine serum (FBS, Gibicco, USA). HT29 cells and HT29-CTX cells were cultured at McCoy’s 5A medium (Solarbio, China) with 10% FBS.

Male Balb/c mice (4 to 6 weeks, 18 to 20 g) were purchased from the experimental animal center of Henan province (Zhengzhou, China). The mice were allowed 10 days to adapt the environment before experiment. All animal experiments were conducted in accordance with the Guidelines for Care and Use of Laboratory Animals of Zhengzhou University and approved by Life Sciences Ethical Review Committee of Zhengzhou University (SYXK-2018-0004, Zhengzhou, China).

**Characterization of particle size and zeta potential:** The nanoparticles (0.1 mg mL<sup>-1</sup>) were dispersed in PBS (pH 7.4) or different physiological fluids for specific time, and then tested the particle size and zeta potential using a Malvern Zetasizer Instrument.

**Optical video recording of nanoparticles and MSD analysis:** The movement of nanoparticles were recorded by inverted microscope with a 100× oil objective. The SiO<sub>2</sub>/Pt and BCTN were dispersed in mucin solution and placed in a glass slide with a cover, and irradiated with 808 nm lasers. The movement of nanoparticles were imaged for 10 s in a bright field. The mean-squared displacement (MSD) were calculated as following (41):

\[
MSD(\Delta t) = [x_i(t + \Delta t) - x_i(t)]^2
\]

$i = 2$ for 2D analysis

**Drug release experiments:** CP@SiO<sub>2</sub>/Pt, CP@SiO<sub>2</sub>/Pt@PEG-Glu were dispersed in PBS (pH 7.4) or GSH containing PBS (5 μM, pH 7.4), and BCTN were dispersed in PBS (pH 7.4), artificial gastric fluid and artificial colonic fluid. At scheduled times, the solutions were centrifuged at...
12000 rpm for 10 min to precipitate the nanoparticles. The content of CP in the supernatant were measured by HPLC.

**Caco-2 and CT26 cellular uptake experiments:** SiO$_2$/Pt, SiO$_2$/Pt@PEG, SiO$_2$/Pt@PEG-Glu, and SiO$_2$/Pt@PEG-Glu@SAM were loaded with FITC. Caco-2 cells were seeded in 12-well plates for 24 hours. Then FITC-loading SiO$_2$/Pt, SiO$_2$/Pt@PEG, and SiO$_2$/Pt@PEG-Glu (50 μg mL$^{-1}$, in terms of SiO$_2$/Pt) were incubated with Caco-2 cells for 3 hours. The cells in SiO$_2$/Pt@PEG-Glu + NIR group received 808 nm laser irradiation (1.5 W cm$^{-2}$) for 5 min after addition of nanoparticles. In addition, dapagliflozin (12.5 μM) was added in the medium of Caco-2 cells in SiO$_2$/Pt@PEG-Glu + dapagliflozin group to inhibit glucose transporter in Caco-2 cells (61). Then the cells were washed with PBS (pH 7.4) and detected by CLSM. The procedure to investigate CT26 cellular uptake was the same as that of Caco-2 cells.

**In vitro biosafety experiments:** The enterotoxin detections of *Staphylococcus aureus* and SAM were carried out by *Staphylococcal* enterotoxin assay kit (RIDASCREEN® SET Total, R-Biopharm AG, Germany) according to commercial instructions. Caco-2 cells were seeded in 96-well plates and incubated for 12 hours. Different concentrations of SiO$_2$/Pt@PEG-Glu, SiO$_2$/Pt@PEG-Glu@SAM were added into 96-well plates and incubated for 24 hours. The cells in SiO$_2$/Pt@PEG-Glu@SAM + NIR group received 808 nm laser irradiation (1.5 W cm$^{-2}$) for 5 min after the addition of nanoparticles. After being washed with PBS (pH 7.4), the cells were incubated in medium containing 10 μL of CCK-8 for 2 hours. The absorbance of each well was detected by microplate reader at 450 nm. The procedure to examine the in vitro biosafety of the nanoparticles on CT26 cells was the same as that of Caco-2 cells.

**Cell viability experiments:** CT26 cells were seeded in 96-well plates and incubated for 12 hours. Different drug concentrations of CP, CP@SiO$_2$/Pt, and CP@SiO$_2$/Pt@PEG-Glu (in terms of CP) were added into 96-well plates and incubated for 24 hours. The cells in SiO$_2$/Pt@PEG-Glu + NIR group received 808 nm laser irradiation (1.5 W cm$^{-2}$) for 5 min after addition of nanoparticles. After being washed with PBS (pH 7.4), the cells were incubated in medium containing 10 μL of CCK-8 for 2 hours. The absorbance of each well was detected by microplate reader at 450 nm.

**Live/dead cell staining experiments:** CT26 cells were seeded in 12-well plates and incubated for 12 hours. CP, CP@SiO$_2$/Pt, and CP@SiO$_2$/Pt@PEG-Glu (5 μg mL$^{-1}$, in terms of CP) were added into 96-well plates and incubated for 24 hours. The cells in SiO$_2$/Pt@PEG-Glu + NIR group received 808 nm laser irradiation (1.5 W cm$^{-2}$) for 5 min after the addition of nanoparticles. The cells in NIR group received different power of 808 nm laser irradiation for 5 min. Then the cells were stained by calcein/PI cell viability assay kit (Beyotime Biotechnology, Shanghai, China) and detected by CLSM.

**In vivo biosafety experiments:** The mice were randomly divided into 4 groups (n = 3), and then received intragastric administration of saline (control) and BCTN (6 mg kg$^{-1}$) twice a week for 3 weeks. Twelve hours after of intragastric administration of saline and BCTN, the mice in NIR and BCTN + NIR groups received 808 nm laser irradiation (1.5 W cm$^{-2}$) for 5 min. The mice in each group and was recorded body weight every 2 days. After 3 weeks of treatment, the mice in each group was sacrificed to take the blood to examined the serum biochemical index and hematological parameters, and the tumor and main tissues were harvested for H&E staining.

**In vivo colocalization of BCTN in gastrointestinal tracts:** SiO$_2$/Pt@PEG-Glu were labeled with FITC, and SAM were labeled with DiI. Then SAM was coated on SiO$_2$/Pt@PEG-Glu to prepare dual fluorescence labeled BCTN. Then the orthotopic tumor-bearing mice received intragastric administration of dual fluorescence labeled BCTN (20 mg kg$^{-1}$). After 3 hours, 6 hours, 12 hours, and 24 hours, the mice were anesthetized and imaged under the small animal imaging
system (IVIS Spectrum, PerkinElmer, USA). After imaging, the mice in each group were sacrificed and the gastrointestinal tract was exteriorized for fluorescent section and the fluorescence was semi-quantitatively analyzed by Image Pro Plus 6.0 Software.

**In vivo biodistribution experiments**: SiO\textsubscript{2}/Pt, SiO\textsubscript{2}/Pt@PEG, SiO\textsubscript{2}/Pt@PEG-Glu, and BCTN were labeled with the near-infrared fluorophore Cy5.5. The orthotopic tumor-bearing mice received intragastric administration of Cy5.5-labeled SiO\textsubscript{2}/Pt, SiO\textsubscript{2}/Pt@PEG, SiO\textsubscript{2}/Pt@PEG-Glu, and BCTN (1 mg kg\textsuperscript{-1}, in terms of Cy5.5). After 6 hours, 12 hours, and 24 hours, the mice received intraperitoneally injection of D-luciferin (150 mg kg\textsuperscript{-1}, J&K Chemical, Ltd., China). Then the mice were anesthetized and imaged under the small animal imaging system (IVIS Spectrum, PerkinElmer, USA). After imaging, the mice in each group were sacrificed and the gastrointestinal tract was exteriorized for fluorescence imaging, fluorescent section. Afterwards, the concentration of Si in different tissues was detected by inductively coupled plasma-mass spectrometry (ICP-MS) after dissolution with aqua regia.

**Blood clearance kinetic experiments**: SiO\textsubscript{2}/Pt, SiO\textsubscript{2}/Pt@PEG-Glu, and SiO\textsubscript{2}/Pt@PEG-Glu@SAM were labeled with Cy5.5. Fourteen days after orthotopic tumor cell inoculation, the mice received intragastric administration of Cy5.5 labeled SiO\textsubscript{2}/Pt, SiO\textsubscript{2}/Pt@PEG-Glu, and SiO\textsubscript{2}/Pt@PEG-Glu@SAM (1 mg kg\textsuperscript{-1}, in terms of Cy5.5). Two hours after intragastric administration, the mice in SiO\textsubscript{2}/Pt@PEG-Glu@SAM + NIR group received 808 nm laser irradiation (1.5 W cm\textsuperscript{-2}) for 5 min. 200 µL of blood was collected from orbital vein to quantify the fluorescence signal of nanoparticles in plasma at specific time intervals (0.5 hour, 1 hour, 2 hours, 4 hours, 6 hours, 8 hours, 12 hours, and 24 hours).

**In vivo degradation experiments**: The orthotopic tumor-bearing mice received intragastric administration of BCTN (3 mg kg\textsuperscript{-1}, in terms of CP) twice a week for 3 weeks. Twelve hours after intragastric administration, the mice received 808 nm laser irradiation (1.5 W cm\textsuperscript{-2}) for 5 min. At 1 day, 15 days and 30 days after 3 weeks of treatment, the mice were sacrificed, then the gastrointestinal tract, main organs, blood, feces and tumors were exteriorized. Afterwards, the concentration of Si in different tissues was detected by ICP-MS after dissolution with aqua regia.

**Long term survival experiments**: Fourteen days after orthotopic tumor cell inoculation, the mice were randomly divided into 3 groups (n = 5), and then received intragastric administration of saline (control), CP (3 mg kg\textsuperscript{-1}), and BCTN (3 mg kg\textsuperscript{-1}, in terms of CP) twice a week for 3 weeks (day 1 to day 21). Twelve hours after intragastric administration, the mice in BCTN + NIR group received 808 nm laser irradiation (1.5 W cm\textsuperscript{-2}) for 5 min. The mice in each group and was recorded body weight every 2 days. Then the mice in each group were continued to observe for one month after treatment to evaluated the long-term survival rate and tumor recurrence possibility (day 22 to day 50). Furthermore, the mice in each group received intraperitoneally injection of D-luciferin (150 mg kg\textsuperscript{-1}, J&K Chemical, Ltd., China) at day 1, day 21 and day 50. Then the mice were anesthetized and imaged under the small animal imaging system (Caliper Life Sciences, Inc., USA) to monitor the tumor bioluminescence. At day 50, the mice in each group was sacrificed to take the tumor and main tissues for bioluminescent imaging to check the tumor cells.

**Statistical Analysis**: Sample sizes and biological replication were determined on the basis of previously published studies and specified in the figure legends. Statistical analysis between groups was analyzed by GraphPad Prism 6.0 software with analysis of variance (ANOVA). * \( P < 0.05 \) is considered a statistical difference; ** \( P < 0.01 \) is considered a significant statistical difference; *** \( P < 0.001 \) is considered an extremely significant statistical difference.
Fig. S1. TGA curves of SiO$_2$/Pt@PEG, SiO$_2$/Pt@PEG-Glu, and SiO$_2$/Pt@PEG-Glu@SAM.
**Fig. S2.** Negative staining TEM image of BCTN.
**Fig. S3.** Particle size and zeta potentials of SAM ($n = 3$). Values are represented as mean ± SD.
Fig. S4. Comparison of the FT-IR spectra of SAM, SiO$_2$/Pt@PEG-Glu and SiO$_2$/Pt@PEG-Glu@SAM.
Fig. S5. Colocalization experiment of SiO$_2$/Pt@PEG-Glu and SAM.
**Fig. S6.** The fluorescence intensity of magnetic beads after different time of incubation with BCTN ($n = 3$). Values are represented as mean ± SD.
**Fig. S7.** Colocalization experiment of SiO$_2$/Pt@PEG-Glu and SAM after being incubated in PBS (pH 7.4), artificial gastric fluid, artificial intestinal fluid and artificial colon fluid for 2 hours.
Fig. S8. Negative staining TEM image of BCTN after being incubated in artificial gastric fluid and artificial intestinal fluid for 2 hours (White outer shell indicated the coating of SAM).
Fig. S9. Colocalization experiment of SiO$_2$/Pt@PEG-Glu and SAM after being incubated with artificial colon fluid for different time.
Fig. S10. Colocalization rate of SiO$_2$/Pt@PEG-Glu and SAM after being incubated with artificial colon fluid for different time ($n = 3$). Values are represented as mean ± SD.
Fig. S11. Drug release rate of BCTN in PBS (pH 7.4), artificial gastric fluid, artificial colon fluid, and under NIR laser irradiation (1.5 W cm$^{-2}$, 5 min, $n = 3$). Values are represented as mean ± SD.
Fig. S12. Fluorescence images of Caco-2 cells after incubation with different FITC-loading nanoparticles for 3 hours.
Fig. S13. Flow cytometry analysis of Caco-2 cells after incubation with different FITC-loading nanoparticles for 3 hours.
Fig. S14. Fluorescence images of CT26 cells after incubation with different FITC-loading nanoparticles for 3 hours.
**Fig. S15.** Flow cytometric analysis of CT26 cells in basal chamber after 3 hours of incubation with different FITC-loading nanoparticles in apical chamber.
Fig. S16. Cell viability of Caco-2 (A) and CT26 (B) cells after incubation with different concentration of SiO$_2$/Pt@PEG-Glu, BCTN, and BCTN + NIR for 24 hours ($n = 9$). Cell viability of Caco-2 (C) and CT26 (D) cells treated with different power densities of laser for 24 hours ($n = 9$). Values are represented as mean ± SD.
Fig. S17. Body weight change of mice received different treatment twice a week for 3 weeks (n = 3). Values are represented as mean ± SD.
Fig. S18. H&E staining images of main organs of mice after different treatment twice a week for 3 weeks. The power of NIR laser irradiation was 1.5 W cm$^{-2}$. Scale bars: 100 μm.
Fig. S19. The levels of TNF-α and IL-6 in the serum of mice after three weeks of different treatment ($n = 3$). Values are represented as mean ± SD.
Fig. S20. Blood biochemical levels of the mice after three weeks of different treatment. Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), Total bilirubin (TBIL), and Total protein (TP) as hepatic function indicators, Blood urea nitrogen (BUN) and Serum creatinine A (CREA) as renal function indicators were measured ($n = 3$). Values are represented as mean ± SD.
Fig. S21. Hematological parameters of the mice after three weeks of different treatment. White blood cells (WBC), Red blood cells (RBC), Hemoglobin (HGB), Hematocrit (HCT), Mean corpuscular volume (MCV) and Mean corpuscular hemoglobin (MCH) ($n = 3$). Values are represented as mean ± SD.
Fig. S22. H&E staining images of tumor sites in tumor-bearing mice.
Fig. S23. Component content of bacterial biomimetic membranes in SiO$_2$/Pt@PEG-Glu@BFM, SiO$_2$/Pt@PEG-Glu@ECM, and BCTN (n = 3). Values are represented as mean ± SD.
Fig. S24. Mean fluorescence intensity of gastrointestinal tracts and tumor sites in tumor-bearing mice after oral administration of different nanoparticles at 12 hours ($n = 3$). Values are represented as mean ± SD.
Fig. S25. Fluorescent images of nanoparticles in different gastrointestinal tracts at 12 hours.
Fig. S26. Median fluorescence intensity (MFI) of nanoparticles in different gastrointestinal tracts at 12 hours ($n = 3$). Values are represented as mean ± SD. ***$P < 0.001$. 
**Fig. S27.** Bioluminescence images of gastrointestinal tract to locate the tumor sites in tumor-bearing mice after oral administration of nanoparticles at different time points in Figure 5C.
Fig. S28. Si element in different tissues after 6 hours (A), 12 hours (B), and 24 hours (C) of oral administration. Results from ex vivo measurements by ICP-MS (n = 3). Values are represented as mean ± SD. Significance was calculated via one-way ANOVA with the Tukey’s post-test. ***$P < 0.001$. 

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**Fig. S28.** Si element in different tissues after 6 hours (A), 12 hours (B), and 24 hours (C) of oral administration. Results from ex vivo measurements by ICP-MS ($n = 3$). Values are represented as mean ± SD. Significance was calculated via one-way ANOVA with the Tukey’s post-test. ***$P < 0.001$. 

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**Fig. S29.** CLSM images of colocalization of cell nucleus (blue) and Ki67 positive cells (red) with BCTN (green) at colorectal cancer sites.
**Fig. S30.** Mean fluorescence intensity of nanoparticles in cancerous cells and noncancerous cells after oral administration of nanoparticles ($n = 5$). Values are represented as mean ± SD. Significance was calculated via one-way ANOVA with the Tukey’s post-test. ***$P < 0.001$.**
Fig. S31. Heatmap of fold change values of routine blood examination between normal mice and tumor-bearing mice after three weeks of treatment (1, normal; 2, saline group; 3, CP group; 4, CP@SiO$_2$/Pt group; 5, CP@SiO$_2$/Pt@PEG-Glu group; 6, BCTN group; 7, BCTN + NIR group, n = 3).
Fig. S32. ALT (A) and AST (B) in the serum of the mice after three weeks of treatment ($n = 3$). Values are represented as mean ± SD. Significance was calculated via one-way ANOVA with the Tukey’s post-test. **$P < 0.01$, ***$P < 0.001$. 
**Fig. S33.** H&E staining images of liver, colorectum and femur in tumor-bearing mice after three weeks of treatment (The blue dotted line indicated the location of tumor metastases).
Fig. S34. Normalized total bone marrow cellularity of mice after three weeks of treatment. The normalized total bone marrow cellularity was determined by ImageJ software to calculate the surface area occupied by total bone marrow cells ($n = 3$). Values are represented as mean ± SD. Significance was calculated via one-way ANOVA with the Tukey’s post-test. $*** P < 0.001$. n.s. means not significant.
**Fig. S35.** Photograph of the tumor dissection harvested in each group after three weeks of treatment (1, saline; 2, CP group; 3, CP@SiO$_2$/Pt group; 4, CP@SiO$_2$/Pt@PEG-Glu group; 5, BCTN group; 6, BCTN + NIR group. Blue circles indicated no identifiable tumor).
**Fig. S36.** Tumor bioluminescence images of mice in each group.
Fig. S37. Survival curve of tumor-bearing mice treated with saline, CP, and BCTN + NIR for three weeks ($n = 5$). Significance was calculated via two-way ANOVA with the Dunnett’s post-test. **$P < 0.01$. 
Fig. S38. Bioluminescence images of main organs of mice after long term survival experiments (H, heart; Li, liver; S, spleen; Lu, lung; K, kidney; LN, Mesenteric draining lymph nodes).
**Fig. S39.** Si element of nanoparticles in different tissues after three weeks of treatment. Results from ex vivo measurements by ICP-MS ($n = 3$). Values are represented as mean ± SD. Significance was calculated via one-way ANOVA with the Tukey’s post-test. ***$P < 0.001$ compared with the same tissue at day 1.
| Formulation       | C<sub>max</sub> (μg/mL) | AUC<sub>0-t</sub> (μg·h/mL) | t<sub>1/2</sub> (h) | F<sub>rel</sub> % |
|-------------------|-------------------------|-----------------------------|---------------------|-----------------|
| SiO<sub>2</sub>/Pt | 0.42±0.08               | 5.44±0.83                   | 6.91±2.85           | 100             |
| SiO<sub>2</sub>/Pt@PEG-Glu | 0.53±0.06             | 7.14±0.83                   | 5.52±0.30           | 131             |
| BCTN              | 0.78±0.06               | 12.01±0.18                  | 9.46±3.74           | 221             |
| BCTN + NIR        | 1.40±0.10               | 19.66±1.16                  | 9.41±0.97           | 361             |

Table S1. Pharmacokinetic parameter of various formulations (n=3).
Legends for videos S1 to S3

**Video S1.** Motion behavior of MSN in mucin solution after 808 nm NIR laser irradiation (1.5 W cm$^{-2}$, 10 s).

**Video S2.** Motion behavior of SiO$_2$/Pt in mucin solution after 808 nm NIR laser irradiation (1.5 W cm$^{-2}$, 10 s).

**Video S3.** Motion behavior of BCTN in mucin solution after 808 nm NIR laser irradiation (1.5 W cm$^{-2}$, 10 s).
REFERENCES AND NOTES

1. R. L. Siegel, K. D. Miller, H. E. Fuchs, A. Jemal, Cancer statistics, 2021. *CA Cancer J. Clin.* **71**, 7–33 (2021).

2. H. Sung, J. Ferlay, R. L. Siegel, M. Laversanne, I. Soerjomataram, A. Jemal, F. Bray, Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J. Clin.* **71**, 209–249 (2021).

3. M. Duran-Lobato, Z. Niu, M. J. Alonso, Oral delivery of biologics for precision medicine. *Adv. Mater.* **32**, 1901935 (2020).

4. S. Hua, Advances in oral drug delivery for regional targeting in the gastrointestinal tract—Influence of physiological, pathophysiological and pharmaceutical factors. *Front. Pharmacol.* **11**, 524 (2020).

5. S. Ahadian, J. A. Finbloom, M. Mofidfar, S. E. Diltemiz, F. Nasrollahi, E. Davoodi, V. Hosseini, I. Mylonaki, S. Sangabathuni, H. Montazerian, K. Fetah, R. Nasiri, M. R. Dokmeci, M. M. Stevens, T. A. Desai, A. Khademhosseini, Micro and nanoscale technologies in oral drug delivery. *Adv. Drug Del. Rev.* **157**, 37–62 (2020).

6. X. Hu, G. Yang, S. Chen, S. Luo, J. Zhang, Biomimetic and bioinspired strategies for oral drug delivery. *Biomater. Sci.* **8**, 1020–1044 (2020).

7. S. P. Bandi, S. Bhatnagar, V. V. K. Venuganti, Advanced materials for drug delivery across mucosal barriers. *Acta Biomater.* **119**, 13–29 (2021).

8. Y. Xu, N. Shrestha, V. Preat, A. Beloqui, Overcoming the intestinal barrier: A look into targeting approaches for improved oral drug delivery systems. *J. Control. Release* **322**, 486–508 (2020).

9. J. T. Huckaby, S. K. Lai, PEGylation for enhancing nanoparticle diffusion in mucus. *Adv. Drug Del. Rev.* **124**, 125–139 (2018).

10. J. Deacon, S. M. Abdelghany, D. J. Quinn, D. Schmid, J. Megaw, R. F. Donnelly, D. S. Jones, A. Kissenpfennig, J. S. Elborn, B. F. Gilmore, C. C. Taggart, C. J. Scott, Antimicrobial efficacy of tobramycin polymeric nanoparticles for Pseudomonas aeruginosa infections in cystic fibrosis:
Formulation, characterisation and functionalisation with dornase alfa (DNase). *J. Control. Release* **198**, 55–61 (2015).

11. H. He, L. Wang, Y. Ma, Y. Yang, Y. Lv, Z. Zhang, J. Qi, X. Dong, W. Zhao, Y. Lu, W. Wu, The biological fate of orally administered mPEG-PDLLA polymeric micelles. *J. Control. Release* **327**, 725–736 (2020).

12. H. C. Zierden, A. Josyula, R. L. Shapiro, H. T. Hsueh, J. Hanes, L. M. Ensign, Avoiding a sticky situation: Bypassing the mucus barrier for improved local drug delivery. *Trends Mol. Med.* **27**, 436–450 (2021).

13. M. Fernández-Medina, M. A. Ramos-Docampo, O. Hovorka, V. Salgueirino, B. Städler, Recent advances in nano- and micromotors. *Adv. Funct. Mater.* **30**, 1908283 (2020).

14. J. Llacer-Wintle, A. Rivas-Dapena, X. Z. Chen, E. Pellicer, B. J. Nelson, J. Puigmart-Luis, S. Pané, Biodegradable small-scale swimmers for biomedical applications. *Adv. Mater.* **33**, e2102049 (2021).

15. X. Zhang, Q. Fu, H. Duan, J. Song, H. Yang, Janus nanoparticles: From fabrication to (bio)applications. *ACS Nano* **15**, 6147–6191 (2021).

16. J. Shao, M. Abdelghani, G. Shen, S. Cao, D. S. Williams, J. C. M. van Hest, Erythrocyte membrane modified janus polymeric motors for thrombus therapy. *ACS Nano* **12**, 4877–4885 (2018).

17. X. Ji, H. Yang, W. Liu, Y. Ma, J. Wu, X. Zong, P. Yuan, X. Chen, C. Yang, X. Li, H. Lin, W. Xue, J. Dai, Multifunctional parachute-like nanomotors for enhanced skin penetration and synergistic antifungal therapy. *ACS Nano* **15**, 14218–14228 (2021).

18. X. Lou, Z. Chen, Z. He, M. Sun, J. Sun, Bacteria-mediated synergistic cancer therapy: Small microbiome has a big hope. *Nano Micro Lett.* **13**, 37 (2021).

19. H. Wu, D. Zhong, Z. Zhang, Y. Li, X. Zhang, Y. Li, Z. Zhang, X. Xu, J. Yang, Z. Gu, Bioinspired artificial tobacco mosaic virus with combined oncolytic properties to completely destroy multidrug-resistant cancer. *Adv. Mater.* **32**, 1904958 (2020).
20. Y. Yang, L. Li, C. Xu, Y. Wang, Z. Wang, M. Chen, Z. Jiang, J. Pan, C. Yang, X. Li, K. Song, J. Yan, W. Xie, X. Wu, Z. Chen, Y. Yuan, S. Zheng, J. Yan, J. Huang, F. Qiu, Cross-talk between the gut microbiota and monocyte-like macrophages mediates an inflammatory response to promote colitis-associated tumourigenesis. *Gut* **70**, 1495–1506 (2021).

21. T. J. Foster, J. A. Geoghegan, V. K. Ganesh, M. Hook, Adhesion, invasion and evasion: The many functions of the surface proteins of *Staphylococcus aureus*. *Nat. Rev. Microbiol.* **12**, 49–62 (2014).

22. K. Kurokawa, K. Takahashi, B. L. Lee, The staphylococcal surface-glycopolymer wall teichoic acid (WTA) is crucial for complement activation and immunological defense against *Staphylococcus aureus* infection. *Immunobiology* **221**, 1091–1101 (2016).

23. P. D. Cani, B. F. Jordan, Gut microbiota-mediated inflammation in obesity: A link with gastrointestinal cancer. *Nat. Rev. Gastro. Hepat.* **15**, 671–682 (2018).

24. Z.-H. Wang, J.-M. Liu, C.-Y. Li, D. Wang, H. Lv, S.-W. Lv, N. Zhao, H. Ma, S. Wang, Bacterial biofilm bioinspired persistent luminescence nanoparticles with gut-oriented drug delivery for colorectal cancer imaging and chemotherapy. *ACS Appl. Mater. Interfaces* **11**, 36409–36419 (2019).

25. M. Wu, Q. Meng, Y. Chen, L. Zhang, M. Li, X. Cai, Y. Li, P. Yu, L. Zhang, J. Shi, Large pore-sized hollow mesoporous organosilica for redox-responsive gene delivery and synergistic cancer chemotherapy. *Adv. Mater.* **28**, 1963–1969 (2016).

26. Z.-H. Wang, J.-M. Liu, N. Zhao, C.-Y. Li, S.-W. Lv, Y. Hu, H. Lv, D. Wang, S. Wang, Cancer cell macrophage membrane camouflaged persistent luminescent nanoparticles for imaging-guided photothermal therapy of colorectal cancer. *ACS Appl. Nano Mater.* **3**, 7105–7118 (2020).

27. H. Armstrong, M. Alipour, R. Valcheva, M. Bording-Jorgensen, J. Jovel, D. Zaidi, P. Shah, Y. Lou, C. Ebeling, A. L. Mason, D. Lafleur, J. Jerasi, G. K. Wong, K. Madsen, M. W. Carroll, H. Q. Huynh, L. A. Dieleman, E. Wine, Host immunoglobulin G selectively identifies pathobionts in pediatric inflammatory bowel diseases. *Microbiome* **7**, 1 (2019).

28. X. Murgia, B. Loretz, O. Hartwig, M. Hittinger, C. M. Lehr, The role of mucus on drug transport and its potential to affect therapeutic outcomes. *Adv. Drug Del. Rev.* **124**, 82–97 (2018).
29. S. B. Wayah, K. Philip, Pentocin MQ1: A novel, broad-spectrum, pore-forming bacteriocin from Lactobacillus pentosus CS2 with quorum sensing regulatory mechanism and biopreservative potential. *Front. Microbiol.* **9**, 564 (2018).

30. J. Kramer, O. Ozkaya, R. Kummerli, Bacterial siderophores in community and host interactions. *Nat. Rev. Microbiol.* **18**, 152–163 (2020).

31. S. A. Torres-Perez, C. E. Torres-Perez, M. Pedraza-Escalona, S. M. Perez-Tapia, E. Ramon-Gallegos, Glycosylated nanoparticles for cancer-targeted drug delivery. *Front. Oncol.* **10**, 605037 (2020).

32. Y. Zhou, Z. Chen, D. Zhao, D. Li, C. He, X. Chen, A pH-triggered self-unpacking capsule containing zwitterionic hydrogel-coated MOF nanoparticles for efficient oral exendin-4 delivery. *Adv. Mater.* **33**, e2102044 (2021).

33. Q. Yan, Y. Lu, L. Zhou, J. Chen, H. Xu, M. Cai, Y. Shi, J. Jiang, W. Xiong, J. Gao, H. Wang, Mechanistic insights into GLUT1 activation and clustering revealed by super-resolution imaging. *Proc. Natl. Acad. Sci. U.S.A.* **115**, 7033–7038 (2018).

34. M. Tutter, C. Schug, K. A. Schmohl, S. Urnauer, C. Kitzberger, N. Schwenk, M. Petrini, C. Zach, S. Ziegler, P. Bartenstein, W. A. Weber, G. Multhoff, E. Wagner, L. H. Lindner, P. J. Nelson, C. Spitzweg, Regional hyperthermia enhances mesenchymal stem cell recruitment to tumor stroma: Implications for mesenchymal stem cell-based tumor therapy. *Mol. Ther.* **29**, 788–803 (2021).

35. P. Icard, S. Shulman, D. Farhat, J.-M. Steyaert, M. Alifano, H. Lincet, How the Warburg effect supports aggressiveness and drug resistance of cancer cells? *Drug Resist. Updat.* **38**, 1–11 (2018).

36. C. Bonner, J. Kerr-Conte, V. Gmyr, G. Queniat, E. Moerman, J. Thévenet, C. Beaucamps, N. Delalleau, I. Popescu, W. J. Malaisse, A. Sener, B. Deprez, A. Abderrahmani, B. Staels, F. Pattou, Inhibition of the glucose transporter SGLT2 with dapagliflozin in pancreatic alpha cells triggers glucagon secretion. *Nat. Med.* **21**, 512–517 (2015).

37. H.-J. Liu, X. Luan, H.-Y. Feng, X. Dong, S.-C. Yang, Z.-J. Chen, Q.-Y. Cai, Q. Lu, Y. Zhang, P. Sun, M. Zhao, H.-Z. Chen, J. F. Lovell, C. Fang, Integrated combination treatment using a “smart”
chemotherapy and microRNA delivery system improves outcomes in an orthotopic colorectal cancer model. *Adv. Funct. Mater.* **28**, 1801118 (2018).

38. Q. Song, C. Zheng, J. Jia, H. Zhao, Q. Feng, H. Zhang, L. Wang, Z. Zhang, Y. Zhang, A probiotic spore-based oral autonomous nanoparticles generator for cancer therapy. *Adv. Mater.* **31**, 1903793 (2019).

39. A. Llopis-Lorente, A. Garcia-Fernandez, E. Lucena-Sanchez, P. Diez, F. Sancenon, R. Villalonga, D. A. Wilson, R. Martinez-Manez, Stimulus-responsive nanomotors based on gated enzyme-powered Janus Au-mesoporous silica nanoparticles for enhanced cargo delivery. *Chem. Commun.* **55**, 13164–13167 (2019).

40. J. Liu, Q. Chen, W. Zhu, X. Yi, Y. Yang, Z. Dong, Z. Liu, Nanoscale-coordination-polymer-shelled manganese dioxide composite nanoparticles: A multistage redox/pH/H₂O₂-responsive cancer theranostic nanoplatform. *Adv. Funct. Mater.* **27**, 1605926 (2017).

41. A. C. Hortelao, R. Carrascosa, N. Murillo-Cremaes, T. Patino, S. Sanchez, Targeting 3D bladder cancer spheroids with urease-powered nanomotors. *ACS Nano* **13**, 429–439 (2019).

42. A. T. Sougiannis, B. N. VanderVeen, R. T. Enos, K. T. Velazquez, J. E. Bader, M. Carson, I. Chatzistamou, M. Walla, M. M. Pena, J. L. Kubinak, M. Nagarkatti, J. A. Carson, E. A. Murphy, Impact of 5 fluorouracil chemotherapy on gut inflammation, functional parameters, and gut microbiota. *Brain Behav. Immun.* **80**, 44–55 (2019).

43. J. Wang, J. Li, J. Yu, H. Zhang, B. Zhang, Large hollow cavity luminous nanoparticles with near-infrared persistent luminescence and tunable sizes for tumor afterglow imaging and chemo-/photodynamic therapies. *ACS Nano* **12**, 4246–4258 (2018).

44. Y. Xiong, Y. Zhao, L. Miao, C. M. Lin, L. Huang, Co-delivery of polymeric metformin and cisplatin by self-assembled core-membrane nanoparticles to treat non-small cell lung cancer. *J. Control. Release* **244**, 63–73 (2016).
45. D.-W. Zheng, X. Dong, P. Pan, K.-W. Chen, J.-X. Fan, S.-X. Cheng, X.-Z. Zhang, Phage-guided modulation of the gut microbiota of mouse models of colorectal cancer augments their responses to chemotherapy. *Nat. Biomed. Eng.* **3**, 717–728 (2019).

46. S. H. Wong, J. Yu, Gut microbiota in colorectal cancer: Mechanisms of action and clinical applications. *Nat. Rev. Gastroenterol. Hepatol.* **16**, 690–704 (2019).

47. K. D. LaCourse, C. D. Johnston, S. Bullman, The relationship between gastrointestinal cancers and the microbiota. *Lancet Gastroenterol. Hepatol.* **6**, 498–509 (2021).

48. J. H. Park, L. Gu, G. von Maltzahn, E. Ruoslahti, S. N. Bhatia, M. J. Sailor, Biodegradable luminescent porous silicon nanoparticles for in vivo applications. *Nat. Mater.* **8**, 331–336 (2009).

49. B. Hu, J. Wang, J. Li, S. Li, H. Li, Superiority of L-tartaric acid modified chiral mesoporous silica nanoparticle as a drug carrier: Structure, wettability, degradation, bio-adhesion and biocompatibility. *Int. J. Nanomedicine* **15**, 601–618 (2020).

50. Q. He, Z. Zhang, F. Gao, Y. Li, J. Shi, In vivo biodistribution and urinary excretion of mesoporous silica nanoparticles: Effects of particle size and PEGylation. *Small* **7**, 271–280 (2011).

51. J. G. Croissant, Y. Fatieiev, N. M. Khashab, Degradability and clearance of silicon, organosilica, silsesquioxane, silica mixed oxide, and mesoporous silica nanoparticles. *Adv. Mater.* **29**, 1604634 (2017).

52. J. Anderski, L. Mahlert, D. Mulac, K. Langer, Mucus-penetrating nanoparticles: Promising drug delivery systems for the photodynamic therapy of intestinal cancer. *Eur. J. Pharm. Biopharm.* **129**, 1–9 (2018).

53. C. Müller, K. Leithner, S. Hauptstein, F. Hintzen, W. Salvenmoser, A. Bernkop-Schnürch, Preparation and characterization of mucus-penetrating papain/poly(acrylic acid) nanoparticles for oral drug delivery applications. *J. Nanopart. Res.* **15**, 1353 (2012).

54. C. Gao, Y. Wang, Z. Ye, Z. Lin, X. Ma, Q. He, Biomedical micro-/nanomotors: From overcoming biological barriers to in vivo imaging. *Adv. Mater.* **33**, e2000512 (2021).
55. M. Wan, Q. Wang, R. Wang, R. Wu, T. Li, D. Fang, Y. Huang, Y. Yu, L. Fang, X. Wang, Y. Zhang, Z. Miao, B. Zhao, F. Wang, C. Mao, Q. Jiang, X. Xu, D. Shi, Platelet-derived porous nanomotor for thrombus therapy. *Sci. Adv.* **6**, eaaz9014 (2020).

56. J. Su, H. Sun, Q. Meng, Q. Yin, S. Tang, P. Zhang, Y. Chen, Z. Zhang, H. Yu, Y. Li, Long circulation red-blood-cell-mimetic nanoparticles with peptide-enhanced tumor penetration for simultaneously inhibiting growth and lung metastasis of breast cancer. *Adv. Funct. Mater.* **26**, 1243–1252 (2016).

57. U. Leung, M. Gonen, P. J. Allen, T. P. Kingham, R. P. DeMatteo, W. R. Jarnagin, M. I. D'Angelica, Colorectal cancer liver metastases and concurrent extrahepatic disease treated with resection. *Ann. Surg.* **265**, 158–165 (2017).

58. D. I. Tsilimigras, P. Brodt, P. A. Clavien, R. J. Muschel, M. I. D'Angelica, I. Endo, R. W. Parks, M. Doyle, E. de Santibanes, T. M. Pawlik, Liver metastases. *Nat. Rev. Dis. Primers.* **7**, 27 (2021).

59. Y. Lu, Q. Zhao, J. Y. Liao, E. Song, Q. Xia, J. Pan, Y. Li, J. Li, B. Zhou, Y. Ye, C. Di, S. Yu, Y. Zeng, S. Su, Complement signals determine opposite effects of B cells in chemotherapy-induced immunity. *Cell* **180**, 1081–1097.e24 (2020).

60. P. Diez, E. Lucena-Sanchez, A. Escudero, A. Llopis-Lorente, R. Villalonga, R. Martinez-Manez, Ultrafast directional Janus Pt-mesoporous silica nanomotors for smart drug delivery. *ACS Nano* **15**, 4467–4480 (2021).

61. E. Vergari, J. G. Knudsen, R. Ramracheya, A. Salehi, Q. Zhang, J. Adam, I. W. Asterholm, A. Benrick, L. J. B. Briant, M. V. Chibalina, F. M. Gribble, A. Hamilton, B. Hastoy, F. Reimann, N. J. G. Rorsman, I. I. Spiliotis, A. Tarasov, Y. Wu, F. M. Ashcroft, P. Rorsman, Insulin inhibits glucagon release by SGLT2-induced stimulation of somatostatin secretion. *Nat. Commun.* **10**, 139 (2019).