**A self-assembling supramolecular dendrimer nanosystem for PET imaging of tumors**

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**Scheme S1:** Synthesis of (A) the Ga-NOTA complex and (B) the dendrimers **1** and Ga-**1**.

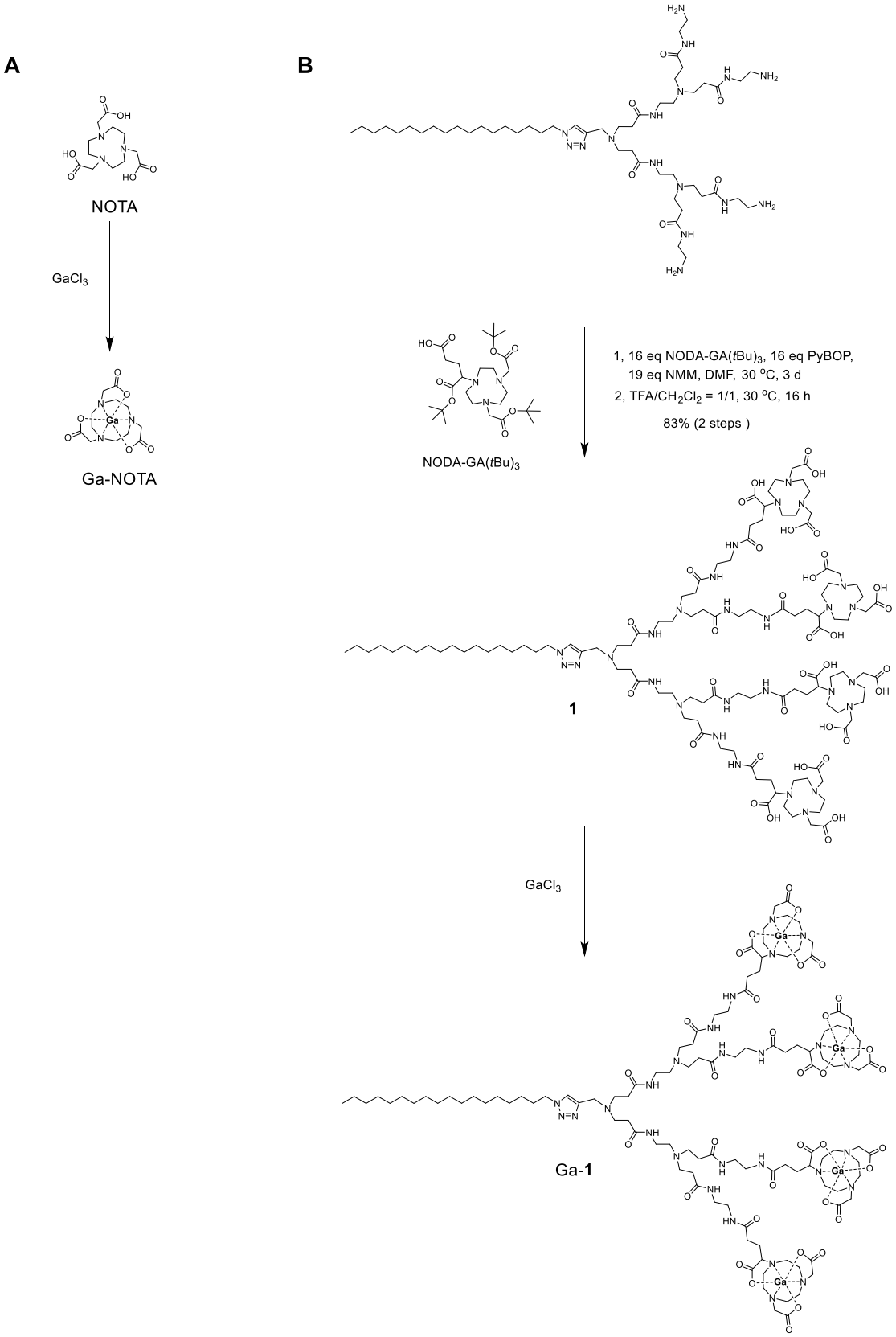
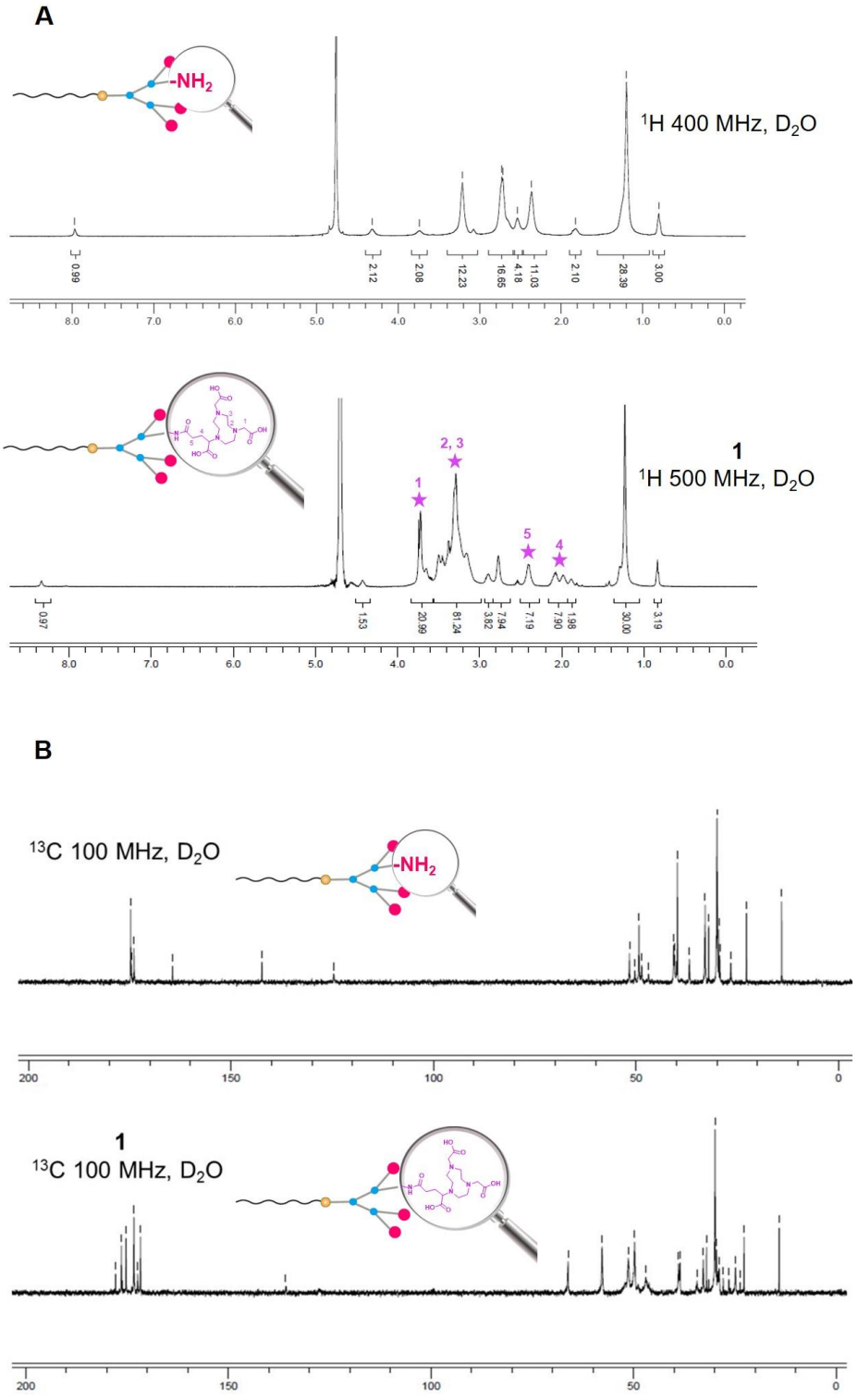
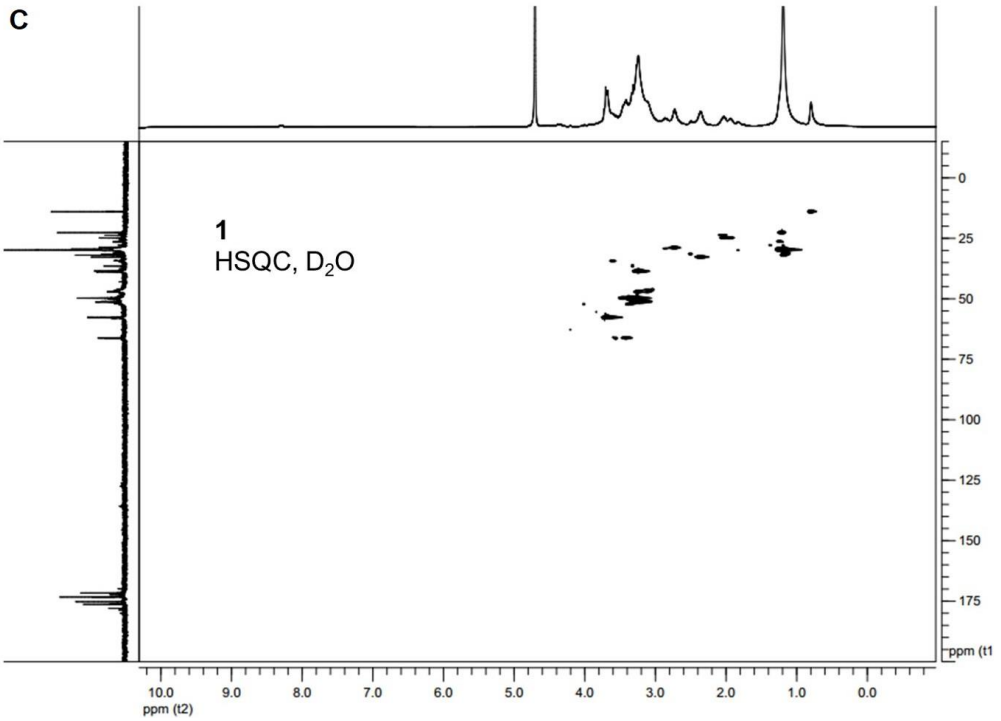


**Figure S1:** (A) $^{1}$H- and (B) $^{13}$C-NMR spectra of the NOTA-conjugated dendrimer **1** compared to the amine-terminated dendrimer recorded in $D_2O$ at 300K. (C) HSQC characterization of NOTA-conjugated dendrimer **1**, and (D) the enlarged region of the HSQC spectrum in (C).

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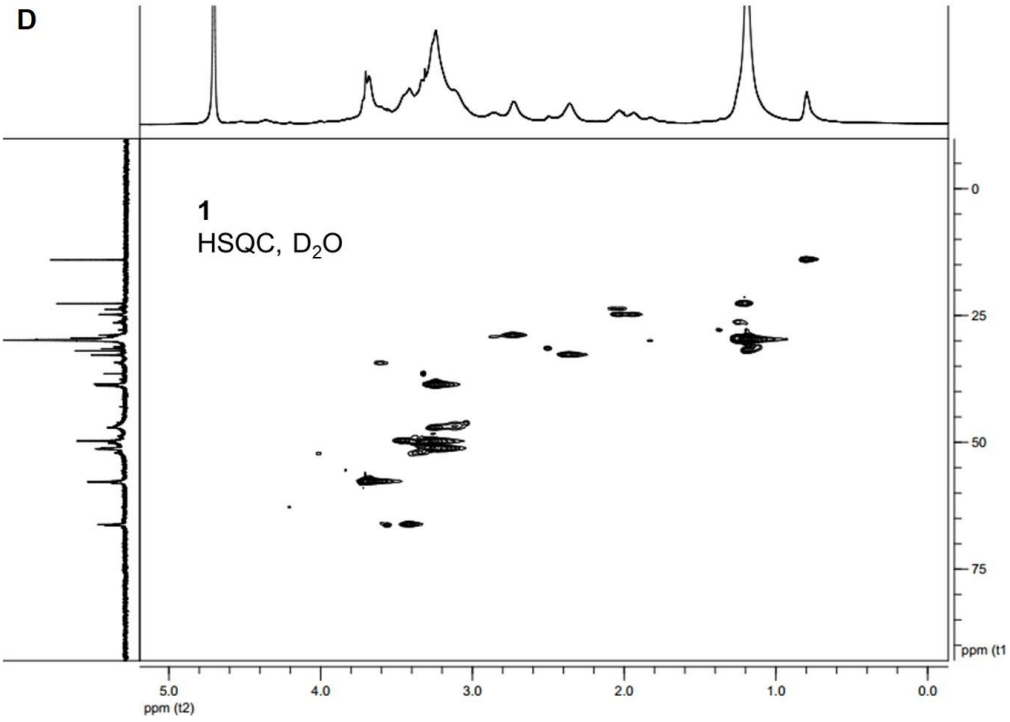
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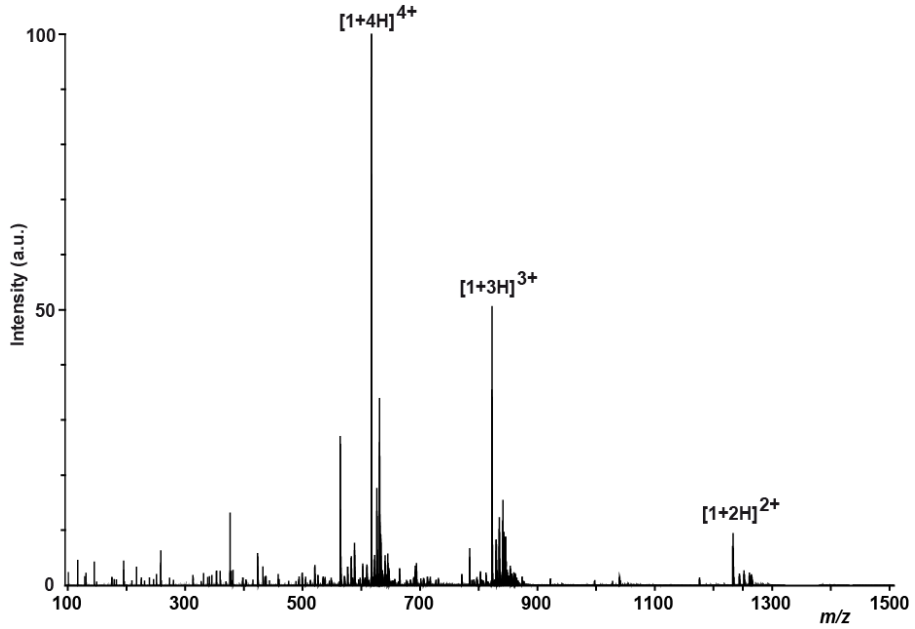
**Figure S2:** ESI-HRMS spectrum of (A) **1** and (B) [$^{69}$Ga]Ga-**1** recorded in acidified methanol.

**A**




**B**

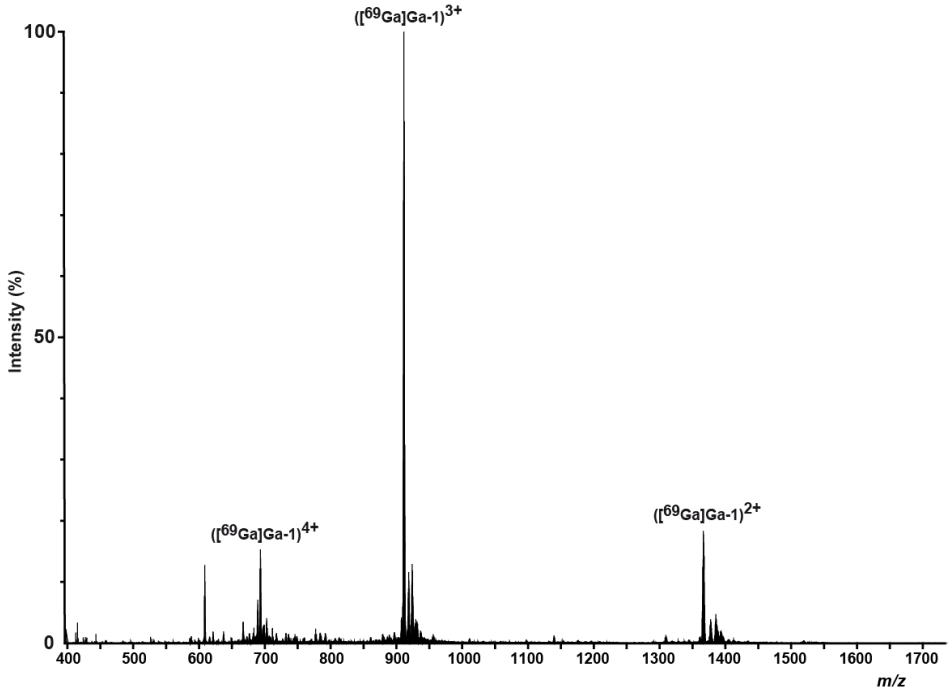


**Figure S3：** The [$^{69}$Ga]Ga-**1** nanosystem is stable with similar size up to one week.

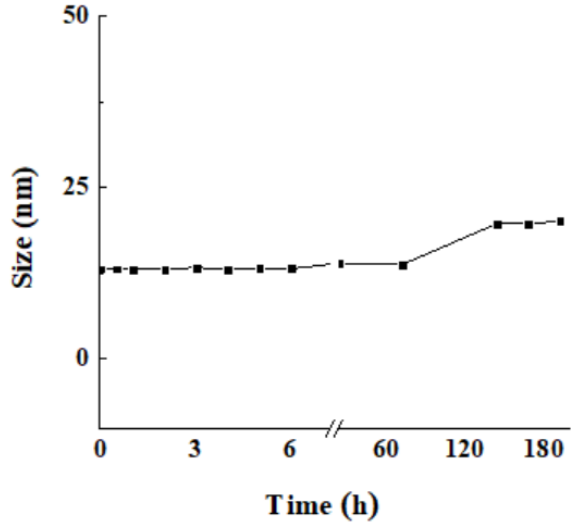


**Figure S4:** (A) Representative radio instant thin-layer chromatogram (radio-iTLC) of [$^{68}$Ga]Ga-**1** without further purification (developing agent: sodium citrate 0.10 M pH = 5.0). (B) Analysis of the radiochemical stability by radio-iTLC of [$^{68}$Ga]Ga-**1** in 0.9% NaCl solution and in human serum at room temperature and at 37°C.

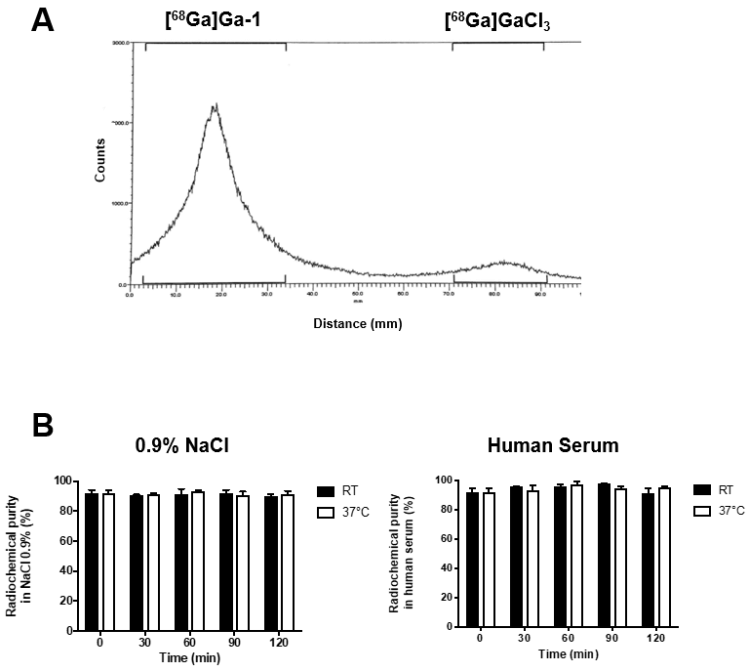


**Figure S5:** Blood clearance of [$^{68}$Ga]Ga-**1** in orthotopic SOJ6-xenograft mice 5, 10, 20, 30, 60 and 90 minutes after intravenous injection. Decay-corrected blood fraction is represented as mean ± SD percentage injected dose per gram for each time point ($n$ = 3 mice).

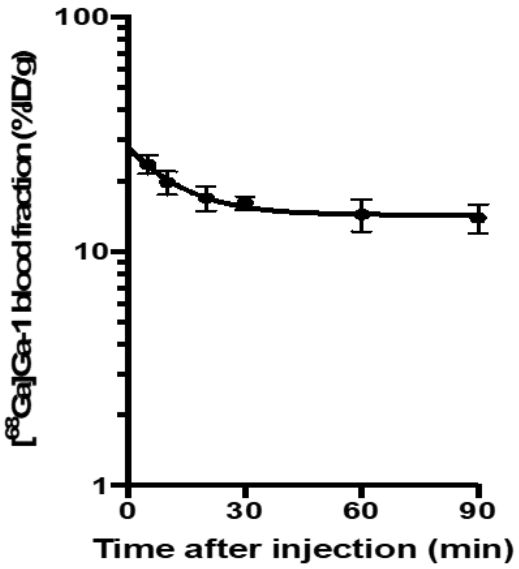


**Figure S6**: Cytokine inducement of **1** and [$^{69}$Ga]Ga-**1** at different doses. 1×, 5× and 10× means the formulations were administered at the dose equal to the PET-imaging dose, 5 times the imaging dose and 10 times the imaging dose, respectively. LPS (lipopolysaccharides), a positive control, was administered *via* intraperitoneal injection at the dose of 5.0 mg/kg. Blood was withdrawn at 3.0 h and 24 h post administration. Concentrations of TNF-α, IFN-γ, IL-6, CXCL1 (KC), MCP-1, GM-CSF, IL-1 and IL-5 were measured with Luminex-based technology. Data revealed that LPS triggered significant cytokine response *in vivo*, however, no inducement of cytokine was observed in mice treated with **1** or [$^{69}$Ga]Ga-**1**, even when they were administered at a dose 10 times higher than the effective PET-imaging dose. Data are shown as mean ± SEM.

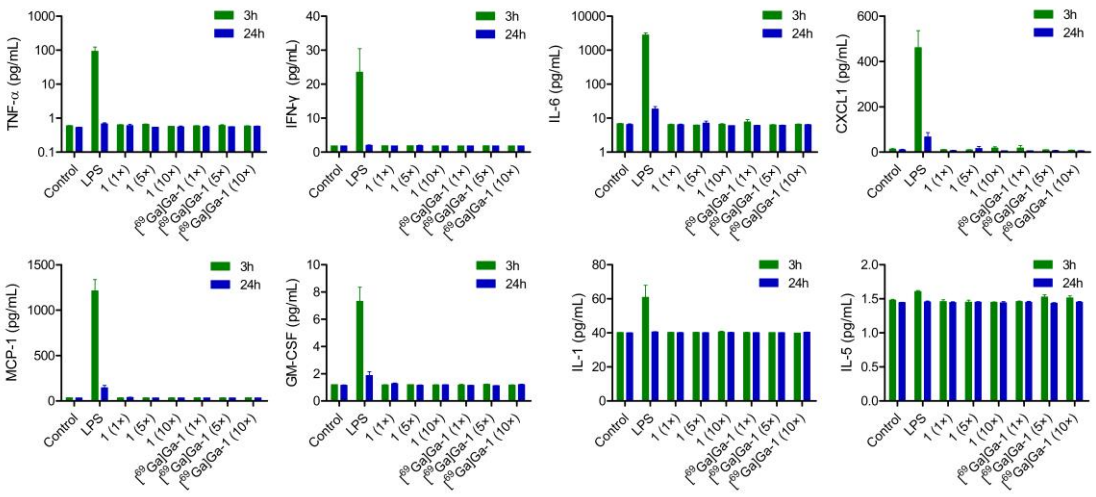


**Figure S7**: Serum biochemistry analysis of mice treated with **1** and [$^{69}$Ga]Ga-**1** at different doses. 1×, 5× and 10× means the formulations were administered at the dose equal to the PET-imaging dose, 5 times the imaging dose and 10 times the imaging dose, respectively. Blood was withdrawn at 3.0 h and 24 h post administration. The major blood biochemistry parameters ALT (alanine transaminase), AST (aspartate transaminase), TBIL (Total Bilirubin), UREA, TP (total protein), ALP (alkaline phosphatase), TCHOL (total cholesterol) and TG (triacylglycerol) remained at the normal level, suggesting that both **1** and [$^{69}$Ga]Ga-**1** are safe *in vivo*, even when administered at a dose 10 times higher than the effective PET-imaging dose. Data are shown as mean ± SEM.

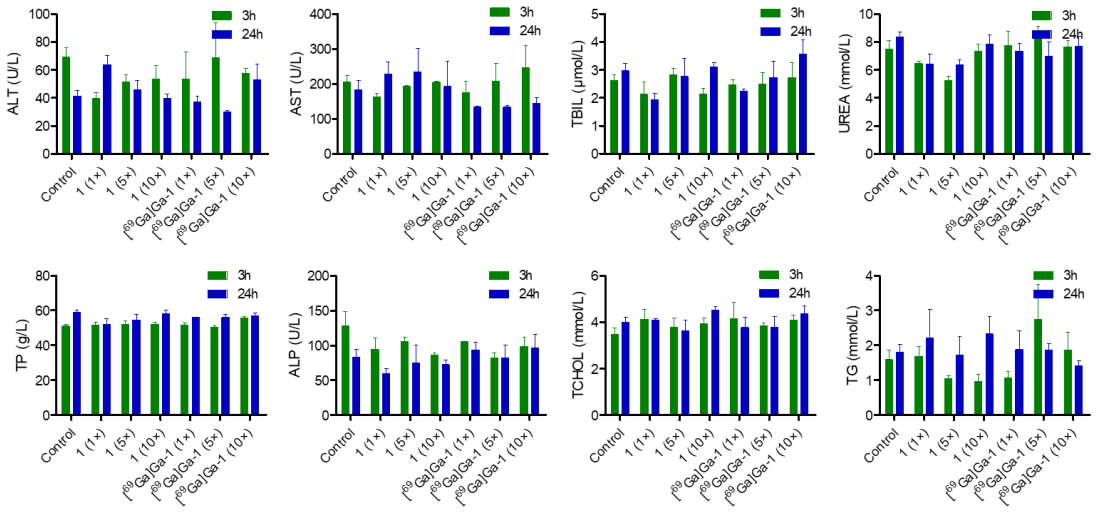


**Figure S8**: Histopathological analysis of main organs from mice treated with **1** and [$^{69}$Ga]Ga-**1** at three different doses: 1× the PET-imaging dose, 5× the imaging dose and 10× the imaging dose. Tissue samples were collected at 24h post administration. No tissue sections revealed any significant histopathological change. Magnification, 200×.

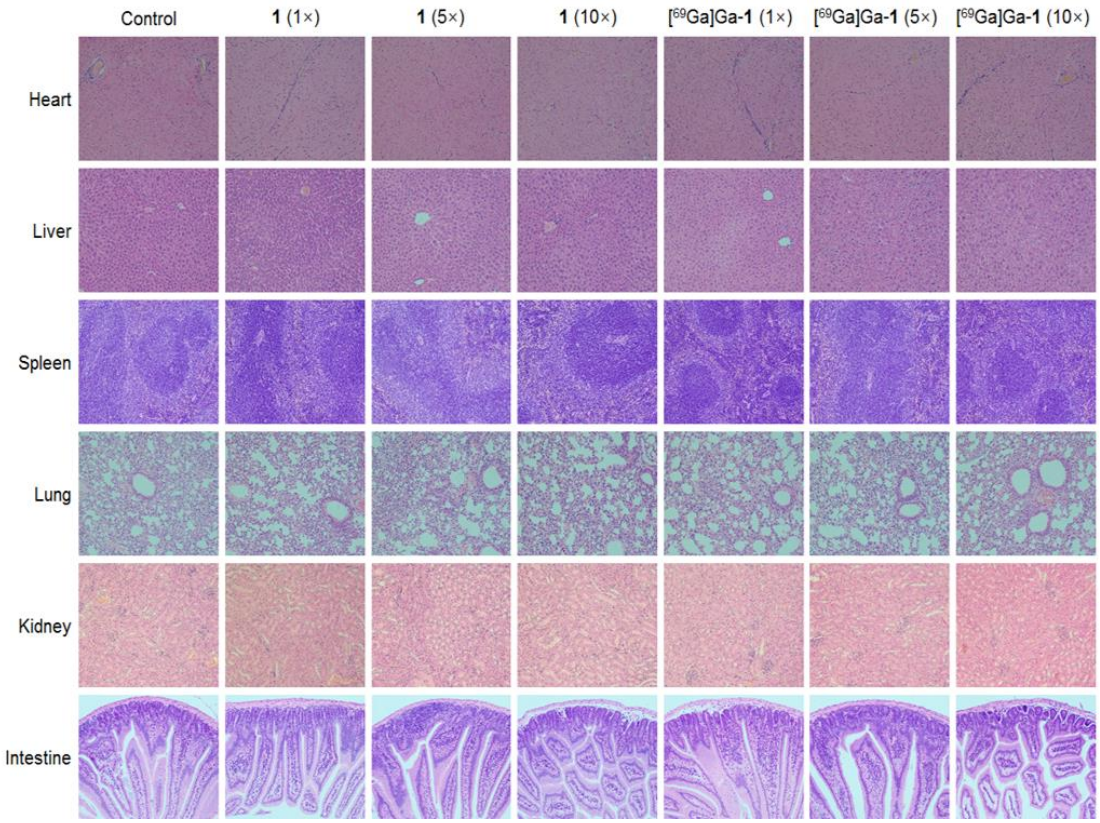


**Table S1 : Injected activities**

| | *n* | **Injected doses (mean ± SD MBq)** | | |
|---|---|---|---|---|
| | | [$^{18}$F]FDG | [$^{68}$Ga]Ga-NOTA | [$^{68}$Ga]Ga-**1** |
| **Tumor model mice** | 3[#] | 6.9 ± 0.2 | 6.4 ± 0.1 | 6.3 ± 1.1 |

[#]for each tumor model.

**General:** The amine-terminated dendrimer **1a** was synthesized according to the well-established protocol published in our group [1-3]. NOTA and NODA-GA($t$Bu)$_3$ was purchased from CheMatech (Dijon, France). Other chemicals were purchased from Acros Organics, Sigma Aldrich or Alfa Aesar. Methyl acrylate, ethylenediamine, trimethylamine, dichloromethane and methanol were dried according to the described methods and distilled before use. The other chemicals were used without further purification. Dialysis tubing was purchased from Sigma Aldrich (St. Quentin Fallavier, France). Analytical thin layer chromatography (TLC) was performed using silica gel 60 $F_{254}$ plates 0.2 mm thick with UV light (254 and 364 nm) as revelator. Radiolabeling analyses were performed on instant thin layer chromatography (iTLC) with a MiniGITA radiochromatography system (Elisia-Raytest, Angleur, Belgium). Chromatography was prepared on silica gel (Merck 200-300 mesh). IR spectra were recorded with an ALPHA FT-IR spectrometer (Bruker, France). $^1$H NMR spectra were recorded at 400 or 500 MHz and $^{13}$C NMR spectra recorded at 100 MHz on Bruker Avance III 400, or JEOL ECS 400 spectrometers. Chemical shifts ($\delta$) are expressed in parts per million (ppm) with the residual peak of $CHCl_3$ at 7.26 ppm or $CH_3OH$ at 3.31 as internal reference. High resolution mass spectrometry experiments were performed with a Synapt G2 HDMS quadrupole/time-of-flight (Manchester, UK) equipped with an electrospray source operating in positive mode. Samples were introduced at 10 µL/min flow rate (capillary voltage +2.8 kV, sampling cone voltage: varied between +20 V and +60 V) under a curtain gas ($N_2$) flow of 100 L/h heated at 35 °C. Accurate mass experiments were performed using reference ions from $CH_3COONa$ internal or external standard. The samples were dissolved and further diluted in methanol (Sigma-Aldrich, St-Louis - MO, USA) doped with formic acid (1% v/v) prior to analysis. Data analyses were conducted using MassLynx 4.1 programs provided by Waters.

**Synthesis and characterization of the amphiphilic dendrimer 1**

To a solution of NODA-GA($t$Bu)$_3$ (0.10 g, 0.18 mmol) in DMF (1.0 mL) were added PyBOP (96 mg, 0.18 mmol) and NMM (22 mg, 0.22 mmol). The mixture was stirred

for 5.0 min and then a solution of the amine-terminating dendrimer (12 mg, 12 μmol) in DMF (1.0 mL) was added and the resulting solution was stirred at 30ºC for 3.0 days under argon. Thereafter, saturated $NaHCO_3$ (15 mL) was added to the solution and ethyl acetate (3.0 × 10 mL) was used for extraction. The combined organic layers were collected, dried over anhydrous $MgSO_4$, filtrated and evaporated under reduced pressure. The crude material was used without any purification for the next step. It was dissolved in a TFA/$CH_2Cl_2$ mixture (3.0 mL, v/v = 1/1) and stirred at 30 ºC for 16 h under argon. After evaporating the solvent, the crude residue was purified by dialysis (dialysis tubing, MWCO 2000) and lyophilized. After repeating 4 times the operation of dialysis and lyophilization, the product was lyophilized to yield the corresponding **1** as a white solid (24 mg, yield: 83%).

$^1$H NMR (500 MHz, $D_2O$): δ 8.34 (s, 1H, -CH- triazole), 4.43 (s, 2H, -$CH_2$-), 3.74-3.65 (m, 22H, -$CH_2$- + -CH-), 3.50-3.16 (m, 84H, -$CH_2$-), 2.89 (s, 4H, -$CH_2$-), 2.89 (s, 8H, -$CH_2$-), 2.41 (br, 8H, -$CH_2$-), 2,09-1.99 (m, 8H, -$CH_2$-), 1.88 (br, 2H, -$CH_2$-), 1.23 (br, 30H, -$CH_2$-), 0.84-0.82 (m, 3H, -$CH_3$); $^{13}$C NMR (100 MHz, $D_2O$): δ176.5, 175.3, 173.4, 172.4, 171.7, 135.8, 127.5, 66.3, 57.8, 51.3, 49.7, 47.2, 47.1, 47.0, 46.9, 46.7, 38.9, 38.5, 36.5, 34.4, 34.3, 32.8, 32.0, 29.9, 29.5, 29.1, 28.8, 27.8, 26.4, 24.8, 22.7, 14.1. HRMS: calcd. for $C_{111}H_{197}N_{28}O_{34}^{3+}$ $[M+3H]^{3+}$ *m/z* 822.4853, measured at *m/z* 822.4860.

**Synthesis and characterization of the amphiphilic dendrimer [$^{69}$Ga]Ga-1**

To the solution of **1** (2.2 μmol, 1.0 mL) was added 90 μL of 2.0 M sodium acetate solution, followed by addition of the solution of 8.9 μmol of [$^{69}$Ga]$GaCl_3$ in 0.50 mL of 1.0 mM HCl. The resulting solution was incubated for 15 min at pH = 4.5 ~ 5.0 at 20 - 25 °C. Then, the solution was subjected to dialysis against water (dialysis tubing, MWCO 2000) for 1 day, then lyophilized to give a white powder [$^{69}$Ga]Ga-**1** (4.7 mg, yield = 85% ). HRMS calcd. for $C_{111}H_{185}N_{28}O_{34}Ga_4^{3+}$ *m/z* 911.3545, measured at *m/z* 911.3547.

**Synthesis and characterization of [$^{69}$Ga]Ga-NOTA**

The preparation of the Ga/NOTA complex followed the similar protocol as for **1**. Briefly,

to 1.5 mL NOTA solution (33 μmol, milliQ water) was added 40 μL of 2.0 M sodium acetate solution, followed by the addition of [$^{69}$Ga]$GaCl_3$ (33 μmol, dissolved in 0.50 mL of 1.0 mM HCl solution). The reaction mixture was incubated for 15 min at pH = 4.5 ~ 5.0 at 20 - 25 °C. The obtained solution was dialyzed against water (dialysis tubing, MWCO 100) for 1.0 day, then lyophilized to give a white powder [$^{69}$Ga]Ga-NOTA (9.9 mg, yield = 81% ). HRMS calcd. for $C_{12}H_{19}N_3O_6Ga^+$ *m/z* 370.0524, measured at *m/z* 370.0522.

**High resolution mass spectrometry**

High resolution mass spectrometry experiments were performed with a Synapt G2 HDMS quadrupole/time-of-flight (Manchester, UK) equipped with an electrospray source operating in positive mode. Samples were introduced at 10 μL/min flow rate (capillary voltage +2.8 kV, sampling cone voltage: varied between +20 V and +60 V) under a curtain gas ($N_2$) flow of 100 L/h heated at 35 °C. Accurate mass experiments were performed using reference ions from $CH_3COONa$ internal standard. The samples were dissolved and further diluted in methanol (Sigma-Aldrich, St-Louis - MO, USA) doped with formic acid (1.0% v/v) prior to analysis. Data analyses were conducted using MassLynx 4.1 programs provided by Waters.

**Critical Micelle Concentration (CMC) measurement**

Critical micelle concentration (CMC) is a well-recognized parameter to evaluate the propensity of an amphiphile to form micellar structures by self-assembly in aqueous solution. Nile red was used as a fluorescence probe to measure CMC values of [$^{69}$Ga]Ga-**1**. Solutions of [$^{69}$Ga]Ga-**1** at different concentrations from $2.0 \times 10^{-7}$ to $1.0 \times 10^{-3}$ mol/L were prepared. The final Nile red concentration was $2.5 \times 10^{-6}$ mol/L in the [$^{69}$Ga]Ga-**1** solutions. Then the solutions were sonicated for 10 min and kept for 2 h at room temperature. Fluorescence spectra were recorded at the excitation wavelength of 550 nm on an F-4500 fluorescence spectrophotometer. The fluorescence intensity at 635 nm was analyzed as a function of dendrimer concentration, and CMC values were

obtained accordingly.

**Dynamic light scattering (DLS)**

Dynamic light scattering (DLS) measurements were performed to determine the hydrodynamic diameter of the nanoparticles formed with the [$^{69}$Ga]Ga-**1** complex. [$^{69}$Ga]Ga-**1** was first dispersed in milliQ water at a concentration of 0.40 mg/mL, and sonicated 30 seconds at 60 Hz (Ultrasonic Cleaner Branson B-200), then the fresh solution was measured using a Malvern Zetasizer Nano ZS equipped with a standard 633 nm laser at 25 °C. The experiments were done in triplicates. The experiments were done in triplicates.

**Transmission Electron Microscopy (TEM)**

Transmission electron microscopy (TEM) was performed using JEOL 3010 transmission electron microscope (Tokyo, Japan) to characterize the size and morphology of the NPs at an accelerating voltage of 300 kV. [$^{69}$Ga]Ga-**1** was dispersed in milliQ water at a concentration of 0.40 mg/mL, and sonicated for 30 seconds, then diluted to 1.6 µg/mL, followed by depositing an aliquot (4.0 µL) onto a carbon-coated copper grid and dried at 37 °C. The grid was then stained with 3.0 µL uranyl acetate (2.0% in aqueous solution) for 4 seconds, and the excess uranyl acetate was removed by filter paper and dried at 37 °C for 60 minutes before measurements.

**Isothermal titration calorimetry (ITC)**

ITC experiments were performed with a MicroCal PEAQ-ITC calorimeter (Malvern, UK) at 25 °C. The cell volume was 208 µL. The binding thermodynamics of monomeric **1** and $Ga^{3+}$ was conducted as follow: a solution of **1** (15 µM in milliQ water ) was titrated with 19 step-by-step injections (2.0 µL each) of a 500 µM solution of $GaCl_3$ in milliQ water. All solutions were degassed for 30 min at room temperature under stirring at 600 rpm prior to each experiment. After careful washing, the cell was pre-rinsed with a portion of milliQ water. Upon filling cell and syringe, stirring was turned on and the system was allowed to thermally equilibrate for 30 minutes.

**Figure S3** shows the titration curve for chelation of $Ga^{3+}$ with the dendrimer **1**. Fitting the curve with a sigmoidal function (red line) yielded the information required to extract the corresponding binding thermodynamics parameters for [$^{69}$Ga]Ga-**1**. Thus, the enthalpy change associated with the cation complexation (ΔH) was determined by subtracting the final (i.e., highest) plateau value from the initial (i.e., lowest) one. The chelation free energy (ΔG) was then calculated from the corresponding $K_a$, obtained as the slope of the sigmoidal curve at its inflection point via the fundamental thermodynamic relationship: $\Delta G = -RT \ln K_a$. $K_d$ was calculated as $1/K_a$. Finally, the entropic variations upon binding were estimated using $\Delta G = \Delta H - T\Delta S$.

**Molecular modeling**

All simulations were carried out using AMBER 18 [4] on a CPU/GPU hybrid cluster. [$^{69}$Ga]Ga-**1** atom types were assigned via the Generalized Amber Force Field (GAFF) [5] and the Visual Force Field Derivation Toolkit (VFFDT) [6]. 100 [$^{69}$Ga]Ga-**1** complexes were randomly placed in a simulation cubic box filled with TIP3 waters [7] and extending at least 2.0 nm from each solute molecule. System neutralization was achieved by adding the appropriate number of the chloride counterions. The hydrated system was subjected to an initial Steepest Descent (SD)/Conjugated Gradient (CG) minimization with 5.0 kcal/(mol$A^2$) restraint on the solute (solvent relaxation), followed by another round of CG minimization without restraints in order to eliminate all bad contacts between water molecules and each solute. Next, the minimized structure was subjected to molecular dynamics (MD) simulations in the canonical (NVT) ensemble under periodic boundary condition. During the 100 ps of MD, each system was gradually heated and relaxed to 25 °C. The SHAKE algorithm [8] was applied to all covalent bonds involving hydrogen atoms. An integration time step of 2.0 fs was adopted together with the Langevin thermostat for temperature regulation (collision frequency = 5.0 $ps^{-1}$) [9]. The final heating step was followed by 100 ns of MD equilibration in the isochoric/isothermal (NPT) ensemble. Pressure control was exerted by coupling the system to a Berendsen barostat (pressure relaxation time 2.0 ps) [8]. The Particle Mesh Ewald (PME) method [10] was used to treat long-range

electrostatic interactions under periodic conditions with a direct space cut-off of 10 Å. Finally, the NPT MD production run was performed for another 1.0 μs. In this case, the Monte Carlo barostat implemented in Amber 18 was adopted for pressure control (1 bar).

**Radiolabeling of [$^{68}$Ga]Ga-1**

Ammonium acetate solution (0.10 mL of 2.0 M) was added to 50 μL dendrimer solution of **1** (1.0 μg/μL in milliQ water). To this solution was added [$^{68}$Ga]$GaCl_3$ solution (0.50 mL) eluted from a $^{68}$Ge/$^{68}$Ga generator (Galliapharm, Eckert&Ziegler, Berlin, Germany). The resulting solution was adjusted to pH = 7.0 and then vortexed and sonicated for 15 minutes at 60 Hz (Ultrasonic Cleaner Branson B-200) to promote the spontaneous self-assembly of [$^{68}$Ga]Ga-**1**. The solution was used for *in vitro* and *in vivo* studies without further purification. The radiochemical purity of 92 ± 2.3% was determined by iTLC (solid phase: iTLC-SG paper purchased from Agilent (Les Ulis, France), mobile phase: 0.10 M sodium citrate pH = 5.0).

**Radiolabeling stability of [$^{68}$Ga]Ga-1**

The radiolabeling stability of [$^{68}$Ga]Ga-**1** was assessed by incubating 0.10 mL of the radiotracer solution in 0.40 mL of 0.90 M sodium chloride solution or in 0.40 mL of human serum respectively. The radiochemical purity stability was checked by iTLC at room temperature right after the radiosynthesis and then at 37°C, 1.0 and 2.0 hours after the radiosynthesis.

**Octanol-water partition coefficient**

To an Eppendorf tube filled with 0.50 mL of the radiolabeled compound [$^{68}$Ga]Ga-**1**, 0.50 mL of octanol was added. The mixture was vigorously stirred by a vortex mixer for 2.0 min at room temperature, then both phases were separated by centrifugation (0.10 kg, 5.0 min). Three 0.10 mL samples were taken from each phase, and measured in a Canberra activimeter, then a mean *log P* was calculated.

**Animals**

All procedures using animals were approved by the Institution's Animal Care and Use Committee (CE14, Aix-Marseille University) and were conducted according to the EU Directive 2010/63/EU and the recommendations of the Helsinki Declaration. Six-week-old BALB/c mice and athymic nude mice were purchased from Envigo. Animals were housed in enriched cages placed in a temperature- and hygrometry-controlled room with daily monitoring, fed with water and commercial diet *ad libitum*.

**Mice ectopic xenograft models of pancreatic, colon, prostate tumors and glioblastoma**

$1.0 \times 10^7$ cells of trypsinized HT29 (human colon adenocarcinoma), 22Rv1 (human prostate cancer), U87 (human glioblastoma), SOJ-6 or L-IPC (primary pancreatic adenocarcinoma) cells were resuspended in 0.50 mL (PBS, Lonza, Basel, Switzerland) with 10% fetal calf serum. Each athymic nude mouse was subcutaneously injected with $5.0 \times 10^6$ tumor cells/0.10 mL between the shoulders. Animals were then allowed for resting during 3.0 weeks.

**Mice orthotopic xenograft models of pancreatic tumors**

$1.0 \times 10^7$ trypsinized SOJ-6 (primary pancreatic adenocarcinoma) cells were resuspended 0.25 mL Roswell Park Memorial Institute (RPMI) 1640 culture media (ThermoFisher Scientific, Waltham, USA) with 10% fetal calf serum and 0.25 mL Matrigel Matrix (Corning, New York, USA). Each athymic nude mouse was injected with $10^6$ SOJ-6 cells/50 µL in the pancreas. Animals were allowed for resting during 3.0 weeks.

**PET biodistribution and tumor uptake study**

For dynamic acquisitions, mice were injected in the caudal vein with [$^{68}$Ga]Ga-**1**, [$^{68}$Ga]Ga-NOTA, or [$^{18}$F]FDG, and immediately imaged for a 2.0 h-dynamic µPET/CT

on a NanoScan PET/CT camera (Mediso, Budapest, Hungary) under 1.5% isoflurane anesthesia. The injected activities are summarized in **Table S1**. Each PET imaging session occurred at a 24 h-delay to prevent any background signal from one imaging session to another. Reconstruction and image treatment (InterView Fusion, Mediso) were carried out to assess tracer uptake in liver, heart, lungs, kidneys, bladder, brain, intestines and spleen, 5.0, 10, 20, 30, 60, 90 and 120 minutes after injection. Results were expressed as mean ± $s_d$ percentage of the injected dose (%ID).

For static acquisitions, 20 min-PET images were acquired respectively 2.0 hours after injection of [$^{68}$Ga]Ga-**1**, 1.0 hour after injection of [$^{68}$Ga]Ga-NOTA, or 40 minutes after [$^{18}$F]FDG injection, on a NanoScan PET/CT camera (Mediso) under 1.5% isoflurane anesthesia. Results were expressed as mean ± $s_d$ percentage of the injected dose per gram of tissue (%ID/g) and as mean±$s_d$ tumor-to-muscle ratio (%).

**Blood kinetics**

Mice were injected in the tail vein with [$^{68}$Ga]Ga-**1**, [$^{68}$Ga]Ga-NOTA, and [$^{18}$F]FDG, respectively, under 1.5% isoflurane anesthesia. The injected activities are summarized in **Table S1**. 10 µL-blood sampling was carried out from the jugular vein at 5.0, 10, 20, 30, 60, and 90 minutes after injection. Samples were then diluted up to 1.0 mL with PBS and counted on a Packard Cobra-II 5002 gamma-counter. Results were decay-corrected, expressed as mean ± $s_d$ percentage of the injected dose per gram of tissue (%ID/g).

**Tumor blood flow measurement**

SOJ-6 orthotopic tumor blood flow was assessed by live 3D-Doppler on a Vevo2100 High Resolution ultrasound system (Visualsonics, Toronto, Canada) with the MS-550D probe (22-55 MHz, axial resolution 40 µm, Visualsonics), by an experienced operator, on 2 time points at a 1-week interval, 3 weeks after xenograft, and 1 hour before [$^{68}$Ga]Ga-1 PET acquisition. Mice were anesthetized using isoflurane 1.5%, secured on a heated animal handling platform enabling electrocardiography, respiration, and temperature monitoring. Ultrasound gel (Aquasonic 100; Parker Laboratories, Fairfield,

NJ, USA) was used as coupling interface between the ultrasound probe and the animal. Tumor blood flow turbulence was measured in a 75 ± 5.0% area from the tumor borders in order to avoid any neighboring vessel likely to interfere with the results.

***In vivo* toxicity**

Animals were maintained in Peking University Laboratory Animal Center (an AAALAC-accredited experimental animal facility). All procedures involving experimental animals were performed in accordance with protocols approved by the Institutional Animal Care and Use Committee of Peking University. Male CD-1 mice 6 - 8 weeks old, weighing 32-38 g, were purchased from Beijing Vital River Laboratory Animal Technology Co., Ltd. Solutions of the dendrimer **1** and the [$^{69}$Ga]Ga-**1** complex were systemically administered into mice *via* tail vein injection (3 animals per group) at three different doses (1× the imaging dose, 5× the imaging dose, and 10 × the imaging dose). Buffer solution and lipopolysaccharide (LPS) were included as control (6 animals). Blood samples were collected and serum specimens were prepared at 3.0 h and 24 h post-administration. A part of serum specimens were sent to Beijing DIAN Clinical Laboratory Co. Ltd., Beijing, China (a subsidiary of Zhejiang DIAN Diagnostics Co., Ltd. China). Concentrations of ALT (alanine transaminase), AST (aspartate transaminase), TBIL (Total Bilirubin), UREA, TP (total protein), ALP (alkaline phosphatase), TG (triacylglycerol) and TCHOL (total cholesterol) in serum were detected with a biochemistry analyzer. Meanwhile, the levels of several cytokines, including TNF-α (tumor necrosis factor alpha), IFN-γ (interferon gamma), IL-6 (interleukin 6), KC (keratinocyte-derived cytokine, or CXCL1, chemokine (C-X-C motif) ligand 1), MCP-1 (monocyte chemoattractant protein-1, or CCL2, chemokine (C-C motif) ligand 2), GM-CSF (granulocyte-macrophage colony-stimulating factor (GM-CSF), or CSF2, colony stimulating factor 2), IL-1 (interleukin 1) and IL-5 (interleukin 5) were measured with immunoassays based on Luminex xMAP (multi-analyte profiling) technology (Beijing 4A Biotech Co., Ltd) according to the manufacturer's protocol. The main organs were also collected at 24 h post-administration, kept and fixed in 10% formalin, paraffin embedded, sectioned and

stained with H&E, and then observed under an optical microscope for histological changes.

**Statistics**

Radiochemical purities were compared with 2-way ANOVA followed by a *post-hoc* Bonferroni test. For each xenografted tumor model, tumor uptakes and tumor-to-muscle ratios were compared with a Wilcoxon matched-pairs signed rank test. Pearson correlation was assessed between PET signal quantification in SOJ-6 orthotopic tumors and the 3D-Doppler-enabled tumor blood flow measurements. Statistical analyses were performed with Prism® software (GraphPad Software). $P < 0.05$ indicated statistical significance.

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