

Supplemental Digital Content 1

LA-ICP-ToF-MS Analysis of Hippocampal Gd Retention In Vivo

For detection of Gd retention inside the hippocampus and dentate gyrus, cryosections of the midbrain (n=2/group and timepoint p.f.i. of GBCAs) were scanned underneath a pulsed beam and ablation spots were lined up edge-to-edge across each line and between consecutive lines without overlapping areas. Laser ablation in the TwoVol3 ablation chamber was carried out in a helium (He) atmosphere using a combined He flow of 0.60 L min⁻¹ and an additional argon (Ar) flow of 0.9 L min⁻¹ injected at the torch of the ICP-ToF-MS. The injection of H₂ and He into the collision/reaction cell (CRC) of the ICP-ToF-MS reduces ⁴⁰Ar⁺, ⁴⁰Ar¹⁶O⁺ and ⁴⁰Ar⁴⁰Ar⁺ interferences (57). A H₂/He gas (8.1 % H₂ and 91.9 % He) was used with a flow rate of approximately 4.0 mL min⁻¹. Instrumental settings were optimised tuning with NIST SRM 610 glass, focusing on high intensities, low oxide formation rates and low elemental fractionation based on ²³⁸U⁺/²³²Th⁺ values close to 1. The effective LA aerosol wash-out time was 10 ms (full-width at 1 percent maximum, FW0.01M) based on the signal from ²⁸Si of individual pulses on NIST SRM 610 glass and from the ⁶³Cu signal of individual pulses on gelatin standards. Gas blank signals were measured before each sample and calibration material, respectively. Gelatin multi-element standards (BIO-logi-CAL standard 2, 48 elements; aemas, Oosterhout, The Netherlands) were used as calibration material following recently established procedures (60) and analysed before and after each sample. The following isotopes were used for data evaluation - ⁵⁶Fe, ⁶³Cu, ⁶⁴Zn and ¹⁵⁸Gd. Since the samples and the gelatin-based standards have a natural isotope abundance, the maps for Gd, Fe, Cu and Zn reflect the quantitative distribution in µg/g in the tissue thin sections.

Table SDC_1. Instrument settings and data acquisition conditions for LA-ICP-ToF-MS imaging

<i>NWRimage Laser Ablation system settings</i>	
Fluence	9 J cm ⁻²
Repetition rate	100 Hz
Beam size	10 µm
Mask shape	Square
Scan velocity	1000 µm s ⁻¹
Helium chamber gas flow	0.30 L min ⁻¹
Helium cup gas flow	0.30 L min ⁻¹
Wash-out time (FW0.01M)	10 ms
<i>icpToF 2R settings</i>	
Radio frequency power	1550 W
Argon cool flow	14 L min ⁻¹
Argon auxiliary flow	0.80 L min ⁻¹
Argon nebulizer flow	0.90 L min ⁻¹
Sampling depth	4 mm
CRC H ₂ /Helium gas flow	4 mL min ⁻¹

Table SDC_2. Results of quantitative analysis of Gd concentrations (µg/g) retained in midbrain slices of EAE or HC mice after treatment with GBCAs detected by LA-ICP-ToF-MS (mean ± standard deviation (SD))

C _{Gd} (µg/g)	timepoint	HC Gadopentetate <i>mean ± SD</i>	EAE Gadopentetate <i>mean ± SD</i>	HC Gadobutrol <i>mean ± SD</i>	EAE Gadobutrol <i>mean ± SD</i>
<i>Whole midbrain slice (n=2)</i>	1d p.f.i.	0.611 ± 0.039	2.461 ± 0.720	0.692 ± 0.502	1.129 ± 1.363
	10d p.f.i.	0.410 ± 0.196	1.604 ± 0.491	0.162 ± 0.024	0.231 ± 0.023
	40d p.f.i.	0.203 ± 0.002	0.528 ± 0.062	0.002 ± 0.002	0.003 ± 0.001
<i>Hippocampus (n=2)</i>	1d p.f.i.	0.687 ± 0.144	2.672 ± 0.592	0.721 ± 0.516	1.323 ± 1.603
	10d p.f.i.	0.394 ± 0.209	1.558 ± 0.373	0.205 ± 0.026	0.268 ± 0.005
	40d p.f.i.	0.156 ± 0.027	0.446 ± 0.102	0.002 ± 0.003	0.003 ± 0.001
<i>Dentate gyrus (n=2)</i>	1d p.f.i.	0.797 ± 0.125	3.114 ± 0.431	0.650 ± 0.460	1.287 ± 1.555
	10d p.f.i.	0.478 ± 0.181	1.510 ± 0.506	0.185 ± 0.028	0.250 ± 0.023
	40d p.f.i.	0.246 ± 0.003	0.494 ± 0.167	0.002 ± 0.002	0.003 ± 0.001

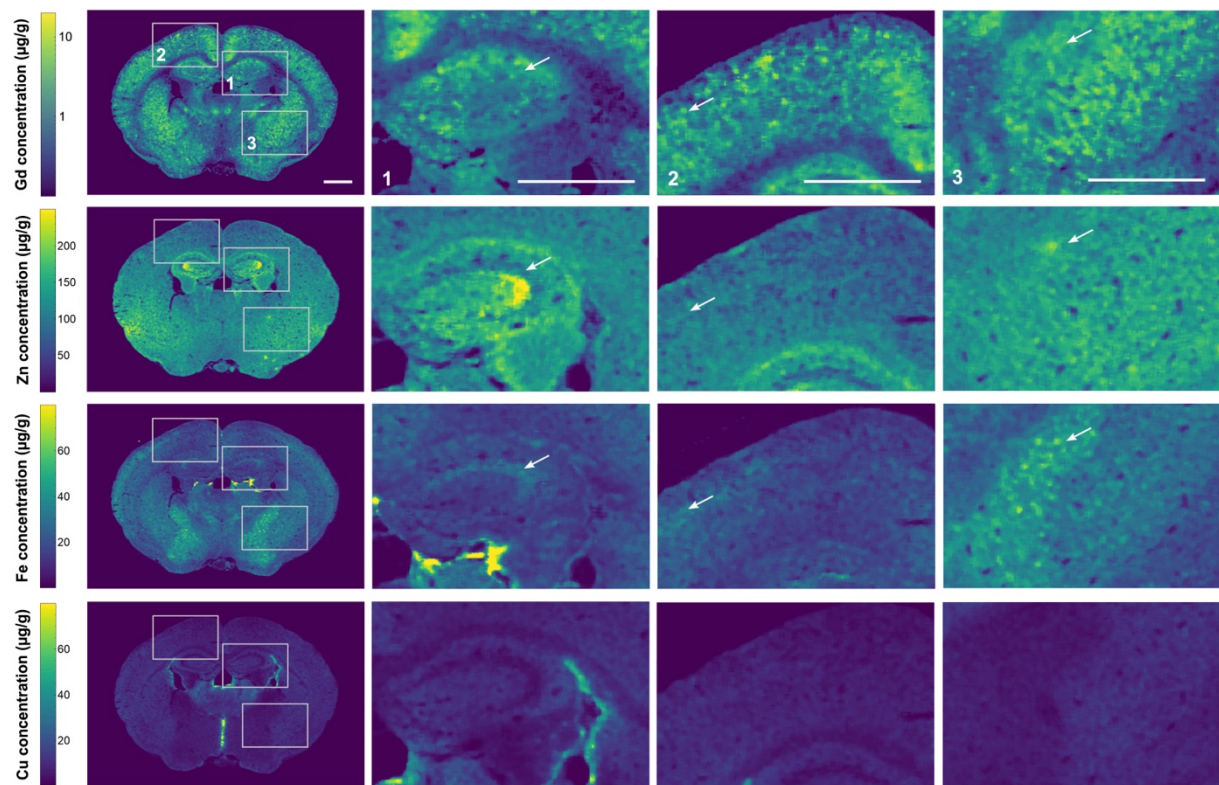


Figure SDC_1. Analysis of elemental distribution patterns of Gd, Fe, Zn and Cu in murine midbrain slice using LA-ICP-ToF-MS following the *in vivo* administration of gadopentetate in an EAE mouse (timepoint: 10 days p.f.i.). Overview maps of the whole midbrain slice are shown on the left, respectively. Zoom-ins for better visualization of regional Gd retention and colocalization with Fe, Zn and Cu are shown on the right for: 1. hippocampus, 2. cortex, 3. globus pallidus and caudoputamen. Gd retention overlap with Zn and Fe distributions inside these three brain regions is indicated with white arrows. *Scale bar: 1 mm.*