SUPPLEMENTARY INFORMATION

Scientific Reports

Neuroprotection by remote ischemic conditioning in the setting of acute ischemic stroke: a preclinical two-centre study

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Supplementary Tables

	London	Lyon
Field strength	9.4T	7T
Gradients	660 mT/m	440 mT/m
Hardware system	Bruker BioSpec	Bruker BioSpec
Software system	ParaVision 6.0.1	ParaVision 5.1
Volume coil for emission: inner diameter (mm)	86	72
Surface coil for reception: diameter (mm)	32	25

Table S1 - MR system characteristics in each center

	T2WI: 2D RARE sequence (RARE factor 8)		**MRA: 2D-inflow angiography with flow compensation**		**DWI: 2D diffusion-weighted echo planar imaging (EPI)**		**PWI: 2D dynamic susceptibility contrast-enhanced (DSC)-MRI**	
MR imaging parameters	**London**	**Lyon**	**London**	**Lyon**	**London**	**Lyon**	**London**	**Lyon**
TE/TR (ms/ms)	75/3500	75/5000	3/18	4.1/18	26.26/5000	23.2/5000	11.1/600	8.2/600
b values (s/mm^2)	N/A	N/A	N/A	N/A	0/1500/3000	0/1500/3000	N/A	N/A
Flip angle (degrees)	180	180	90	90	90	90	25	50
Number of averages	2	2	2	3	2	2	1	1
Bandwidth (kHz)	37	35	100	50	300	300	300	247
Field of view (mm^2)	35 x 35	35 x 35	35 x 31	35 x 25	35 x 35	35 x 35	35 x 35	35 x 35
Slice thickness/interslice (mm)	1/0	1/0	0.4/0.15	0.4/0.25	1/0	1/0	1/0	1/0
Number of slices	15	15	60	60	15	15	15	15
Matrix size	256 x 256	256 x 256	256 x 226	256 x 184	128 x 128	128 x 128	80 x 80	80 x 80
Number of repetitions	N/A	N/A	N/A	N/A	N/A	N/A	100	100
Contrast agent CA (i.v., bolus)	N/A	N/A	N/A	N/A	N/A	N/A	Gd-DO3A-butrol	Gd-DOTA
Dose (ml/kg)	N/A	N/A	N/A	N/A	N/A	N/A	0.6	0.6
Acquisition time (min, sec)	3 min 44 sec	4 min 44 sec	8 min 8 sec	6 min 27 sec	4 min 40 sec	4 min 40 sec	1 min	1 min

Table S2 - MR imaging protocol characteristics in each center

TE: echo time; TR: repetition time; T2WI: T2-weighted MRI; MRA: magnetic resonance angiography; DWI: diffusion-weighted imaging; PWI: perfusion-weighted imaging; N/A: not applicable; Gd-DO3A-butrol: Gadovist (Bayer Healthcare, Germany); Gd-DOTA: DOTAREM (Guerbet, France).

Sign	Description	Score
Motility, **spontanous activity**	Normal	0
	Slightly reduced exploratory behaviour	1
	Moving limbs without proceeding	2
	Moving only to stimuli	3
	Unresponsive to stimuli, normal muscle tone	4
	Premortal signs, severe hypotonia	5
Gait	Straight walking	0
	Walking toward controlateral side	1
	Alternate circling & straight walk	2
	Alternate circling & walking toward paretic side	3
	Circling/other gait disturbances	4
	Constant circling toward paretic side	5
Postural signs	Degree of forelimb flexion when held by tail	0-2
	Degree of body rotation when held by tail	0-2
Parachute reflex	Symmetrical	0
	Asymmetrical	1
	Controlateral forelimb retracted	2
Lateral resistance	Degree of resistance against lateral push	0-2
Limb placing	Ipsilateral forelimb: normal, weak, no placing	0-2
	Controlateral forelimb: normal, weak, no placing	0-2
TOTAL		**0-22**

Table S3 - Neuroscore rating

Supplementary Figures and Figure Legends

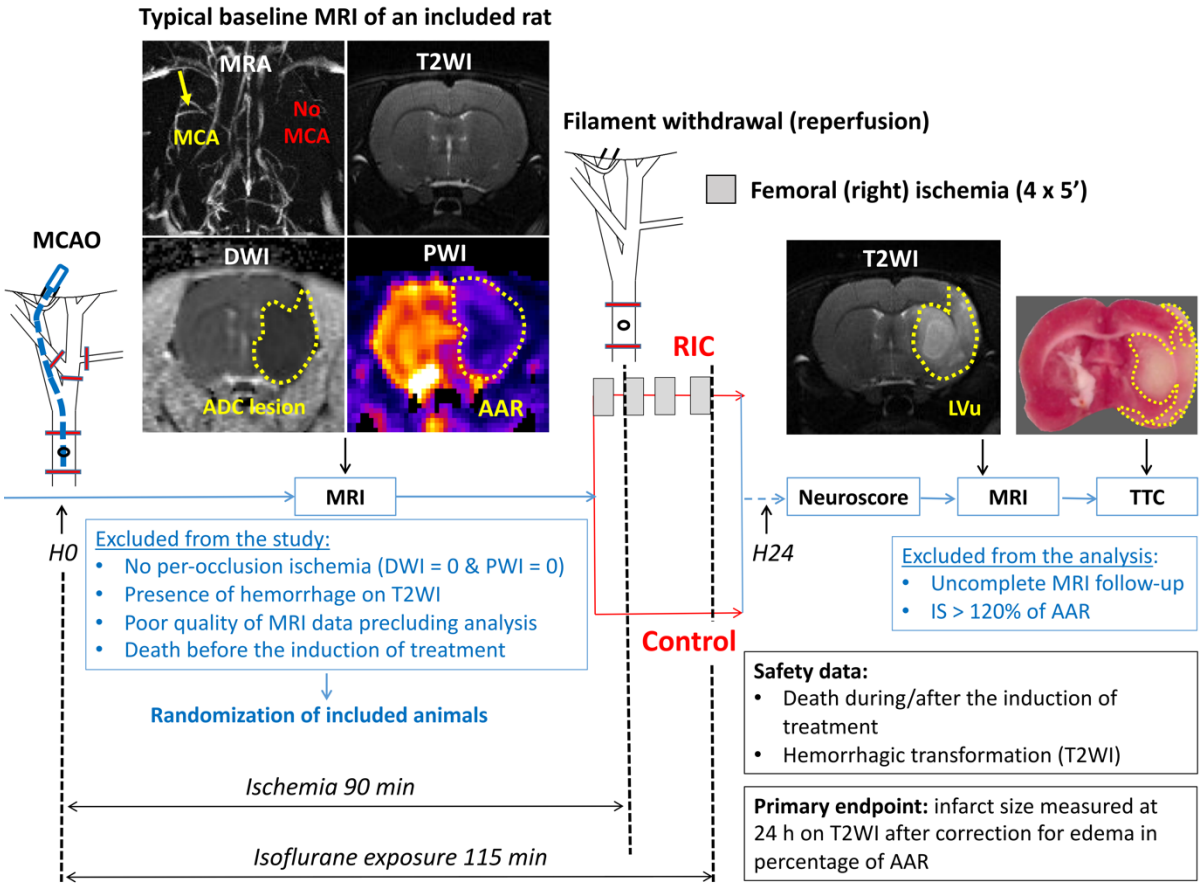

Figure S1 - Study design.

AAR: area at risk; ADC: apparent diffusion coefficient; DWI: diffusion-weighted imaging; H0: baseline time point; H24: 24 hours; IS: infarct size; LVu: lesion volume uncorrected; MCA: middle cerebral artery; MCAO: middle cerebral artery occlusion; MRA: magnetic resonance angiography; MRI: magnetic resonance imaging; PWI: perfusion-weighted imaging; RIC: remote ischemic conditioning; TTC: triphenyl tetrazolium chloride; T2WI: T2-weighted imaging.

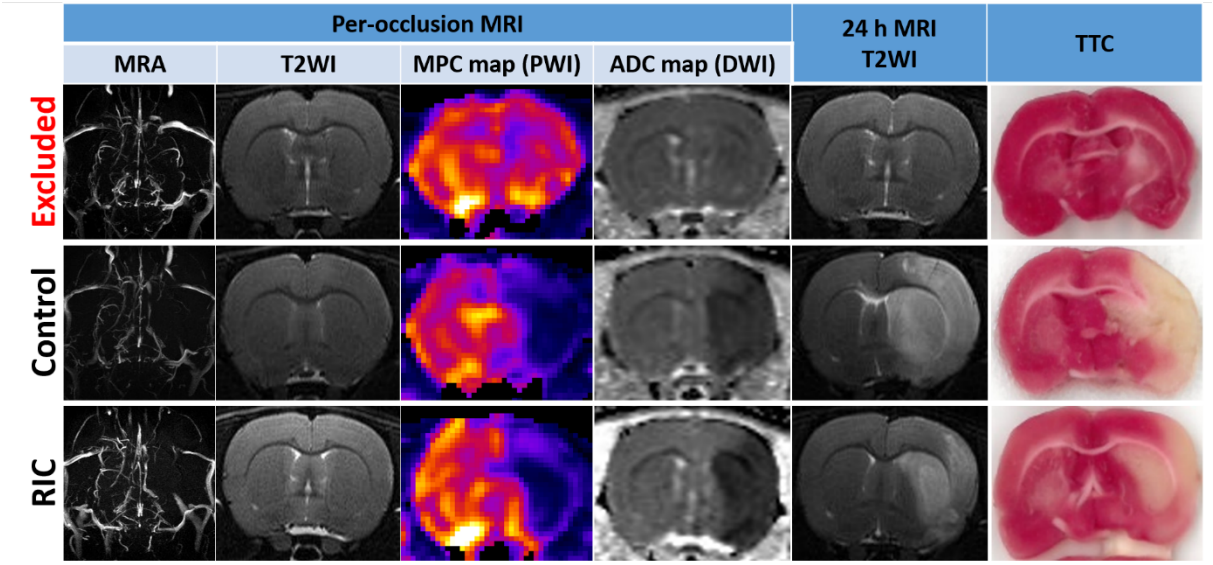

Figure S2 - MRI and TTC data of representative rats.

Only one slice is shown from the 3D data sets. First row: one animal illustrating the inclusion/exclusion criteria: here both PWI and DWI are negative, meaning there is no cerebral ischemia. The MRA score was 2 (indicative of ipsilateral middle cerebral artery patency). This rat was wrongly included at admission and then excluded from the analysis (as shown on the CONSORT-like diagram). As expected, no infarct was present on follow-up T2WI and TTC staining. Second and third rows: animals from the control and RIC groups respectively. These two rats presented with the same per-occlusion MRI features: MRA score of 0 (indicative of total MCA occlusion), AAR of 40% (control) and 50% (RIC) of the hemisphere, corticostriatal ADC lesion of 34% (both control and RIC). At 24h, the infarct has grown up at the expense of the penumbra in the control animal (50%), while it remained within the ADC lesion borders in the animal treated with RIC (29%).

ADC: apparent diffusion coefficient; DWI: diffusion-weighted imaging; MPC: maximum peak concentration; MRA: magnetic resonance angiography; MRI: magnetic resonance imaging; PWI: perfusion-weighted MRI; RIC: remote ischemic conditioning; TTC: triphenyl tetrazolium chloride; T2WI: T2-weighted imaging.

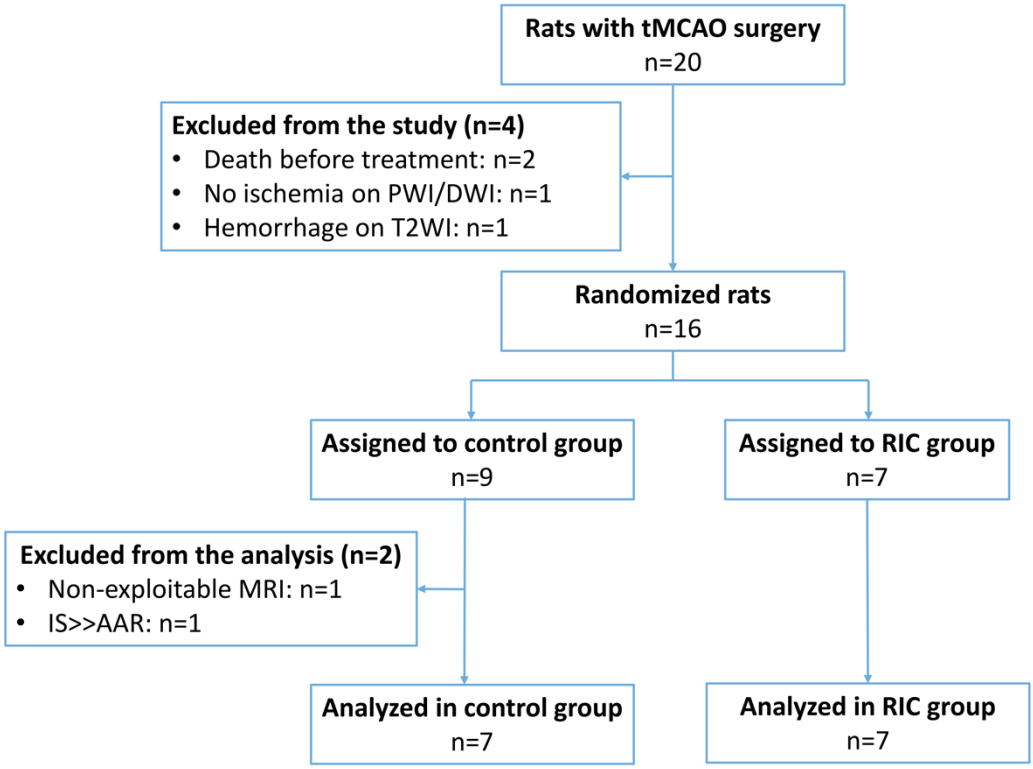

Figure S3 - CONSORT-like diagram of London study.

CONSORT = Consolidated Standards of Reporting Trials; tMCAO: transient middle cerebral artery occlusion; PWI: perfusion-weighted imaging; DWI: diffusion-weighted imaging; T2WI: T2-weighted imaging; RIC: remote ischemic conditioning; MRI: magnetic resonance imaging; IS: infarct size; AAR: area at risk.

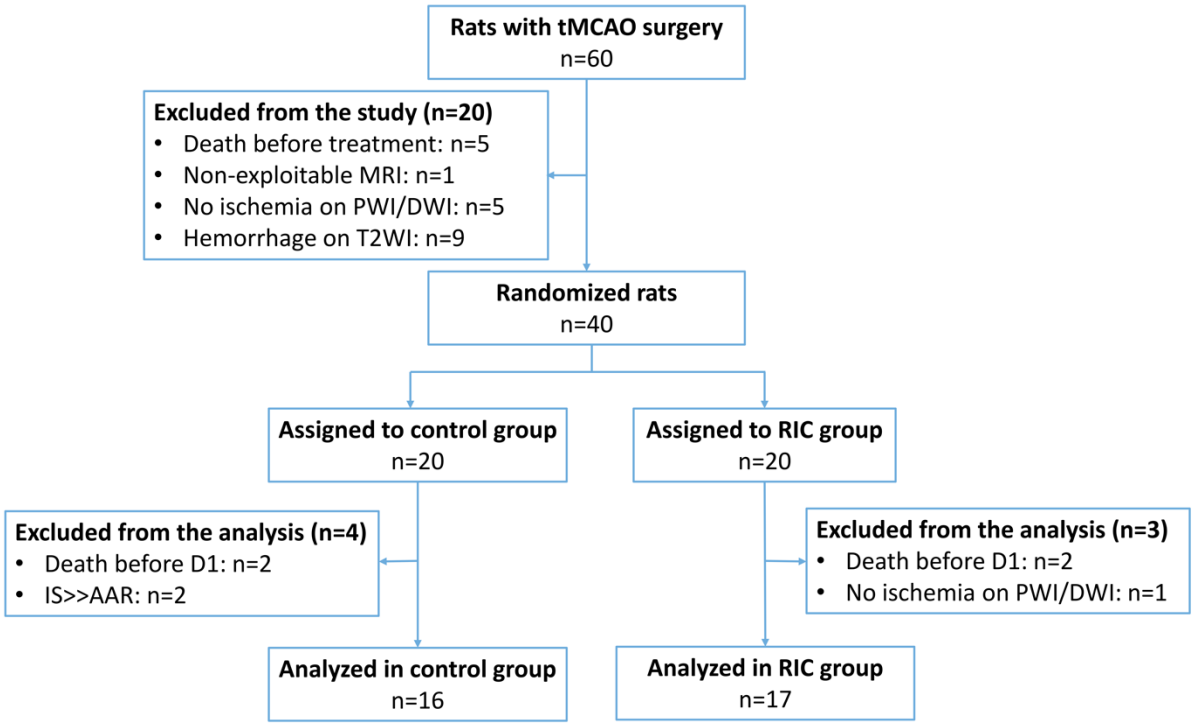

Figure S4 - CONSORT-like diagram of Lyon study.

CONSORT = Consolidated Standards of Reporting Trials; tMCAO: transient middle cerebral artery occlusion; MRI: magnetic resonance imaging; PWI: perfusion-weighted imaging; DWI: diffusion-weighted imaging; T2WI: T2-weighted imaging; RIC: remote ischemic conditioning; IS: infarct size; AAR: area at risk; D1: day 1 post-tMCAO.