

Materials List for:

Two-photon Imaging of Microglial Processes' Attraction Toward ATP or Serotonin in Acute Brain Slices

Fanny Etienne^{1,2,3}, Vincenzo Mastrolia^{1,2,3}, Luc Maroteaux^{1,2,3}, Jean-Antoine Girault^{1,2,3}, Nicolas Gervasi^{*1,2,3}, Anne Roumier^{*1,2,3}

¹Institut national de la santé et de la recherche médicale (INSERM), Institut du Fer-à-Moulin (UMR-S 839)

²Sorbonne Université

³Institut du Fer à Moulin

* These authors contributed equally

Correspondence to: Nicolas Gervasi at nicolas.gervasi@inserm.fr, Anne Roumier at anne.roumier@inserm.fr

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Materials

Name	Company	Catalog Number	Comments
for pipettes preparation			
Clark Borosilicate Thin Wall Capillaries	Harvard Apparatus	30-0065	Borosilicate Thin Wall without Filament, 1.5 mm OD, 1.17 mm ID, 75 mm L , Pkg. of 225
DMZ Universal Puller	Zeitz Instrumente		
Name	Company	Catalog Number	Comments
for solutions			
Calcium Chloride dihydrate (CaCl ₂)	Sigma	C5080	
Choline Chloride	Sigma	C7527	
D-(+)-Glucose	Sigma	G8270	
L-Ascorbic acid	Sigma	A5960	
Magnesium Chloride solution 1M (MgCl ₂)	Sigma	63020	
Potassium chloride SigmaUltra>99,0% (KCl)	Sigma	P9333	
Sodium bicarbonate (NaHCO ₃)	Sigma	S5761	
Sodium Chloride (NaCl)	Sigma	S5886	
Sodium phosphate monobasic	Sigma	S5011	
Sodium pyruvate	Sigma	P2256	
Ultrapure water	MilliQ		for all the solutions
Name	Company	Catalog Number	Comments
for slice preparation			
2x 200 mL crystalizing dishes			
80 mL Pyrex beaker			
Antlia-3C Digital Peristaltic pump	DD Biolab	178961	For mice perfusion and 2-photon chamber perfusion (aCSF)
Carbogen 5% CO ₂ /95% O ₂	Air Liquide France Industrie		
Dolethal	Vetoquinol	Dolethal 50 mg/mL	
Filter papers (Whatman)	Sigma	WHA1001042	Whatman qualitative filter paper, Grade 1 (Pore size: 11µM)
Fine Scissors - Sharp	Fine Science Tools	14060-60	
Food box 10 cm diameter, 8 cm Height			

glue (ethyl cyanoacrylate)	Loctite	super glue 3 power flex	
Hippocampal Tool (spatula)	Fine Science Tools	10099-15	The largest extremity has to be angled at 90 °
Ice			
Iris Forceps (curved)	Moria	MC31	
Lens cleaning tissue	THOR LABS		
Nylon mesh strainer			diameter 7 cm
Razor blades	Electron Microscopy Sciences	72000	For the slicer
scalpel blade			
Slice interface holder			home-made, the file for 3D printing is provided in Supplemental Material
Surgical Scissors - Sharp	Fine Science Tools	14002-14	
Vibrating slicer	Thermo Scientific	720-2709	Model: HM 650V (Vibrating blade microtome)
Water bath			Set at 32°C (first recovery step)
Name	Company	Catalog Number	Comments
for slice imaging			
× 25 0.95 NA water-immersion objective	Leica Microsystems (Germany)	HXC Irapo	
2-photon MP5 upright microscope with resonant scanners (8 kHz) and two HyD Hybrid detectors	Leica Microsystems (Germany)		
Antlia-3C Digital Peristaltic pump	DD Biolab	178961	For 2-photon chamber perfusion with aCSF
Carbogen 5% CO2/95% O2	Air Liquide France Industrie	I1501L50R2A001	
Chameleon Ultra2 Ti:sapphire laser	Coherent (Germany)		
disposable transfer pipettes , wide mouth	ThermoFischer scientific	for example : 232-11	5.8ml with fin tip, but we cut it (approx 7cm) to have a 4 mm diameter mouth
emission filter SP680	Leica Microsystems (Germany)		
fluorescent cube containing a 525/50 emission filter and a 560 dichroic filter (for fluorescence collection)	Leica Microsystems (Germany)		
glass beaker with 50 mL of ACSF to maintain constant perfusion of the slice			
Heating system	Warner Instrument Corporation	Automatic Heater Controller TC-324B	to maintain perfusion solution at 32°C
perfusion chamber			home-made, the file for 3D printing is provided in Supplemental Material
slice holder ("harp")			home made : hairpin made of platinum with the two branches joined by parallel nylon threads
Name	Company	Catalog Number	Comments
for slice stimulation			
Adenosine 5'-triphosphate disodium salt hydrate (ATP)	Sigma	A-26209	to be prepared ex-temporaneously : 1mg/ml (3mM) stock solution prepared the day of the experiment, kept at 4°C (a few hours) and diluted just before use
Fluorescein (optional)	Sigma	F-6377	use at 1 µM final

Micromanipulator	Luigs and Neumann	SM7	connected to the micropipette holde
Micropipette holder			same as for eletrophysiology
Serotonin hydrochloride	Sigma	H-9523	aliquots of 50mM stock solution in H2O kept at -20°C. 500µM solution prepared the day of the experiment.
Syringe 5mL (without needle)	Terumo medical products	SS+05S1	
Transparent tubing	Fischer Scientific	11750105	Saint Gobain Performance Plastics™ Tygon™ E-3603 Non-DEHP Tubing
Name	Company	Catalog Number	Comments
for image analysis			
Fiji		https://fiji.sc	Schindelin, J. et al Nat. Methods (2012) doi 10.1038
Icy	Institut Pasteur	http://icy.bioimageanalysis.org	de Chaumont, F. et al. Nat. Methods (2012)
Name	Company	Catalog Number	Comments
mice			
CX3CR1-GFP mice	Jung et al, 2000		male or females, P3 to 2 months-old ; we have backcrossed these mice on 129sv background.
CX3CR1creER-YFP mice	Parkhurst et al 2013		male or females, P3 to 2 months-old ; we have backcrossed these mice on 129sv background.