

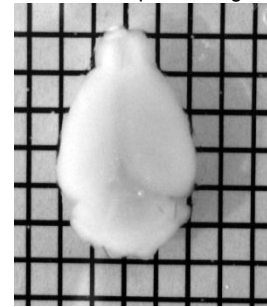
VA-044

For Transparent Tissue Imaging

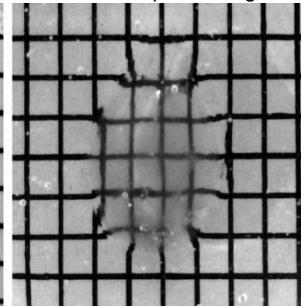
CLARITY is the technique developed in the Deisseroth lab at Stanford University.
Chung, K. *et al.*, Nature, 497(7449):332-337 (2013).
This protocol transforms intact tissues including whole organs into an optically transparent hydrogel-hybridized form, ready for molecular phenotyping and high resolution 3-D imaging. FUJIFILM Wako's VA-044 is used to initiate hydrogel polymerization.

Note: Data was provided by FUJIFILM Wako Pure Chemical Corporation

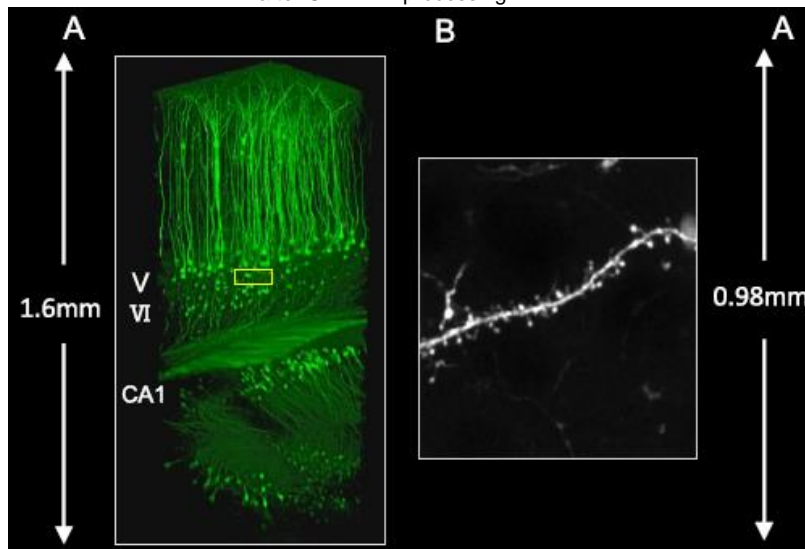
Mouse Brain BEFORE CLARITY processing



Mouse Brain AFTER CLARITY processing

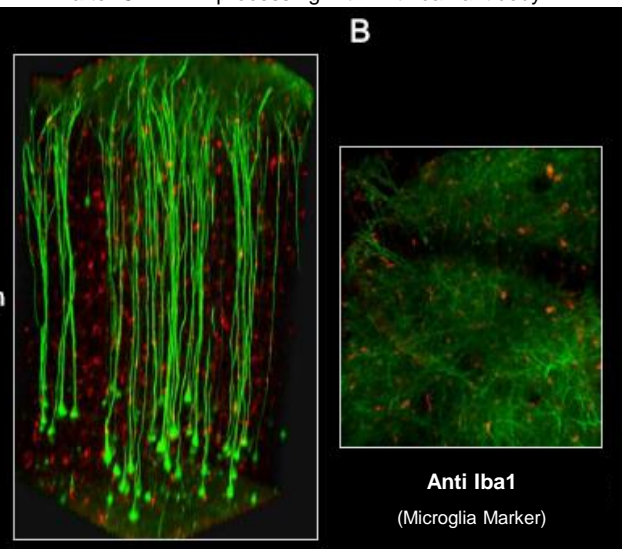


Thy1-YFP (H Line) Mouse Brain Imaging after CLARITY processing



- A. 3D imaging of Cerebral Cortex to Hippocampus
 - B. Imaging of cone cells dendrite of Cerebral Cortex Layer V
- Photographing condition : 2Photon
Olympus FV1000 upright microscope with 25times liquid immersion objective lens

Thy1-YFP Mouse Brain Imaging after CLARITY processing with Anti Iba1 antibody



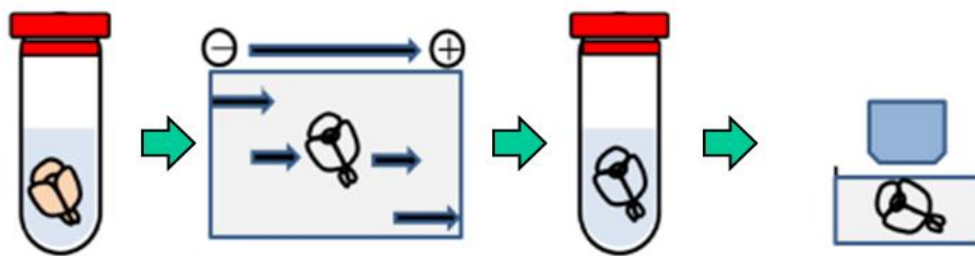
- A. 3D imaging of Microglia Cell in Cerebral Cortex
 - B. Imaging of surface of Cortex Layer I
- Photographing condition : 1Photon
Olympus FV1000 upright microscope with 25times liquid immersion objective lens

VA-044 for Transparent Tissue Imaging

Catalog No.	Product Name	Grade	Storage	Unit
LB-VA044-50GS	VA-044	For Cellbiology	Keep at RT	50 g
LB-VA044-500GS	2,2'-Azobis[2-(2-imidazolin-2-yl)propane]dihydrochloride			500 g
019-19741	Anti Iba1, Rabbit	Immunocytochemistry	-20C	50 µg (100 µL)
631-26271	Electrophoresis Chamber for Lipid Extraction (Nihon Eido)	-	-	1 machine

**The product is intended for laboratory research use only and not to be used for drug, food or human use.

CLARITY Protocol



Hydrogel Monomer
Infusion under 4°C (days 1-3)
Hydrogel-tissue hybridization
under 37°C (day 3)

Electrophoretic tissue cleaning
around 37°C (days 2-5)
2 days for Glycerol immersion
fluorescent protein

Immunostaining with
antibody (days 2-5)

2 days for Glycerol immersion
for Observation with 2 photon
microscope

[Hydrogel Monomer Solution Components]

▪ VA-044 -----	1 gram
▪ 40% Acrylamide -----	40 mL
▪ 2% BisAcrylamide -----	10 mL
▪ 10 x PBS -----	40 mL
▪ 16% Paraformaldehyde ---	100 mL
▪ Saponin -----	200 mg
▪ Water -----	210 mL

[Electrophoresis Buffer]

▪ Boric Acid -----	123.66 gram
▪ SDS -----	400 gram
▪ NaOH -----	Adjust to pH 8.5
▪ Water -----	Dilute to 10 L

[Electrophoresis Chamber for Lipid Extraction]

[Circulation Pump]

Catalog No.	CLARITY Reagents	Grade	Storage	Unit
017-08012	Acrylamide	For Electrophoresis	Keep at RT	25 g
019-08011				100 g
011-08015				500 g
138-06032	N,N'-Methylenebis (acrylamide)	For Electrophoresis	Keep 2-10°C	25 g
130-06031				100 g
163-25265	10 x PBS(-)	For Cell Culture	Below 25°C	500 mL
160-16061	Paraformaldehyde	For Tissue Fixation	Keep at RT	100 g
162-16065				500 g
163-25983	16w/v% Paraformaldehyde Solution , Methanol free	For Electron Microscopy	Keep at RT	10 mL x 10A
193-18651	Saponin, from Soybeans	Wako 1 st Grade	Keep 2-10°C	1 g
199-18653				5 g
029-02191	Boric Acid	JIS Special Grade	Keep at RT	100 g
021-02195				500 g
190-13982	Sodium Dodecyl Sulfate	For Molecular Biology	Keep at RT	25 g
192-13981				100 g
194-13985				500 g

References for CLARITY– from brain to the whole body

Kadoki, M. *et al.*, Cell, 171(2):398-413.e21 (2017)
 Ku, T. *et al.*, Nature Biotechnology, 34(9):973-981 (2016)
 Ye, L. *et al.*, Cell, 165(7):1776-1788 (2016)
 Yang, B. *et al.*, Cell, 158(4):945-958 (2014)

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