

3D Imaging Kit for Mouse Brain

Dream technology :
optimized solution for clearing tissue without damage

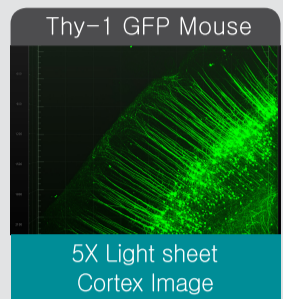
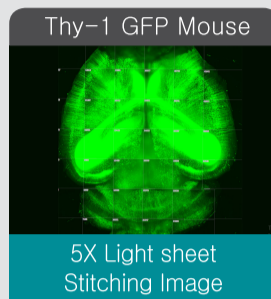
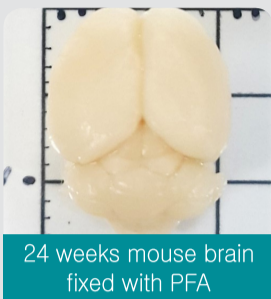
✓ **NO NEED DEDICATED EQUIPMENT & PREPROCESSING**
GET IMAGE WITH JUST SHAKING



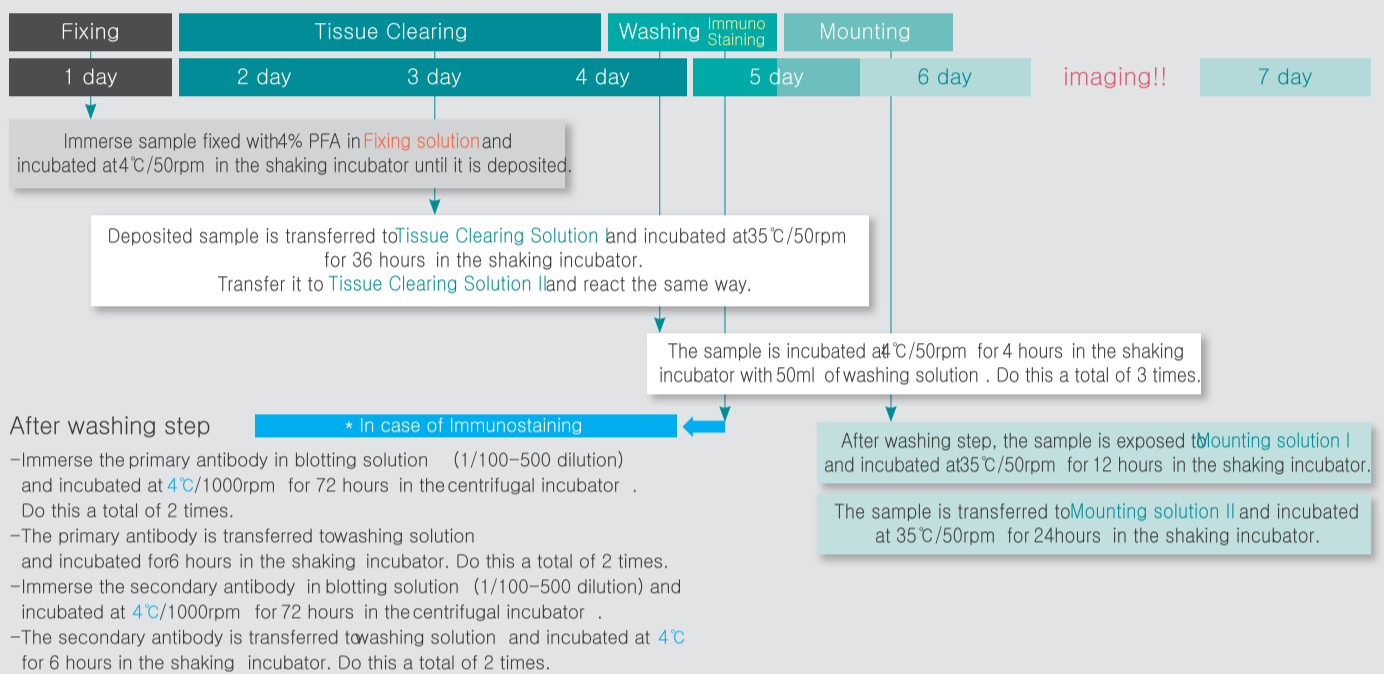
KIT
Components

1	Fixing Solution	40 mL
2	Tissue Clearing Solution I	6 mL
3	Tissue Clearing Solution II	6 mL
4	Washing Solution	400 mL
5	Blotting Solution	20 mL
6	Mounting Solution I	10 mL
7	Mounting Solution II	10 mL
8	Image Chamber	1 ea

✓ **A WEEK FOR ALL CLEARING AND IMAGING**



Start!



✓ **Protocol**

★ Preparation

Pre-incubation of Tissue Clearing Solution (I, II) and Mounting Solution (I, II) at 35 °C for 1 hour to clear bottom of bottles.
Fixation of transcardial perfusion sample at 4 °C over 12 hours using 4 % Paraformaldehyde (PFA) solution

★ Tissue Clearing

- Put 4% PFA fixed sample in the **Fixing solution** and incubate at 4 °C, 50 rpm shaker to be deposited.
- Put deposited sample in **Tissue Clearing Solution I** and incubate at 35 °C, 50 rpm shaker for 36 hours. After the first incubation repeat this step in **Tissue Clearing Solution II**.*
- Add 50 ml of **Washing Solution** to sample and incubate at 4 °C, 50 rpm shaker for 4 hours. Repeat this step 3 times.*
- Put washed sample in **Mounting Solution I** and incubate at 35 °C, 50 rpm shaker for 12 hours.
- After mounting, **centrifuge** sample at 1200 rpm for 30 minutes and remove **bubbles**.
- Put sample in **Mounting Solution II** and incubate at 35 °C, 50 rpm shaker for 24 hours.
- Put transparent sample in Image Chamber and close a lid avoiding bubble.
- After checking image, sample can be stored in image chamber or 1X PBS at 4 °C

★ Cautions

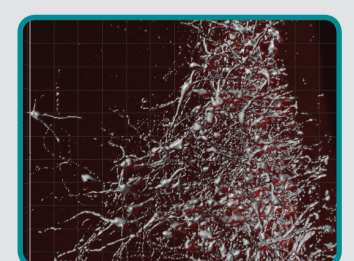
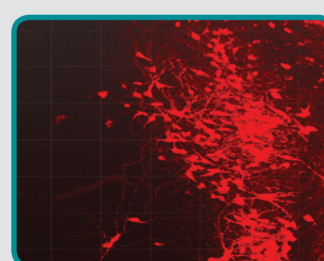
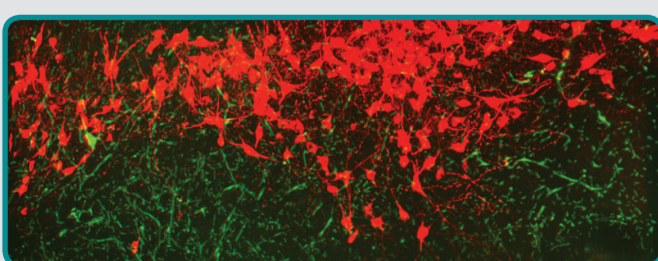
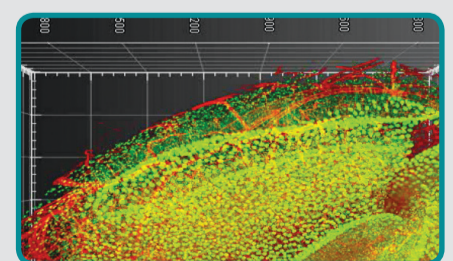
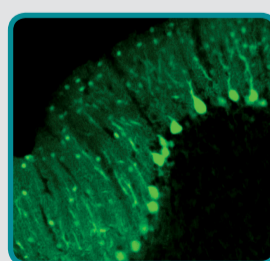
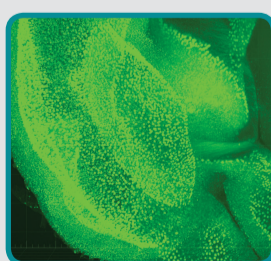
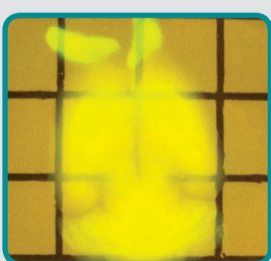
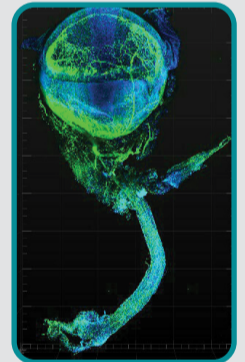
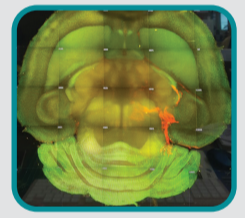
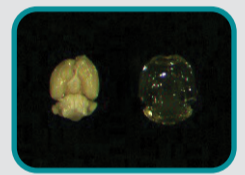
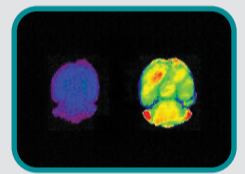
Kit must be stored at 4 °C
Opened bottle must be used within 1 month

- *Step 2, in case of whole brain Axon bundle can be found but proceed to washing step.
- *Step 3, if step 2 and 3 are repeated again you can achieve more transparent sample.

★ Immuno staining

After washing,

- Centrifugal incubation at 4 °C, 1000 rpm for 3 days in 1:100~1:500 primary antibody diluted with Blotting Solution. Repeat this step twice.
- Add washing solution to sample and shake it for 6 hours. Repeat this step twice.
- Centrifugal incubation at 4 °C, 1000 rpm for 3 days in 1:100~1:500 secondary antibody diluted with Blotting Solution.
- Add washing solution and shake at 4 °C for 6 hours. Repeat this step twice.
- Continue the beginning of step 4



Ordering information

Product	Code Number
SunHyun 3D Imaging kit	SH3DI-0101

