Product Data Sheet

IS-0012 Fluorojade C Stain (1.000 X)

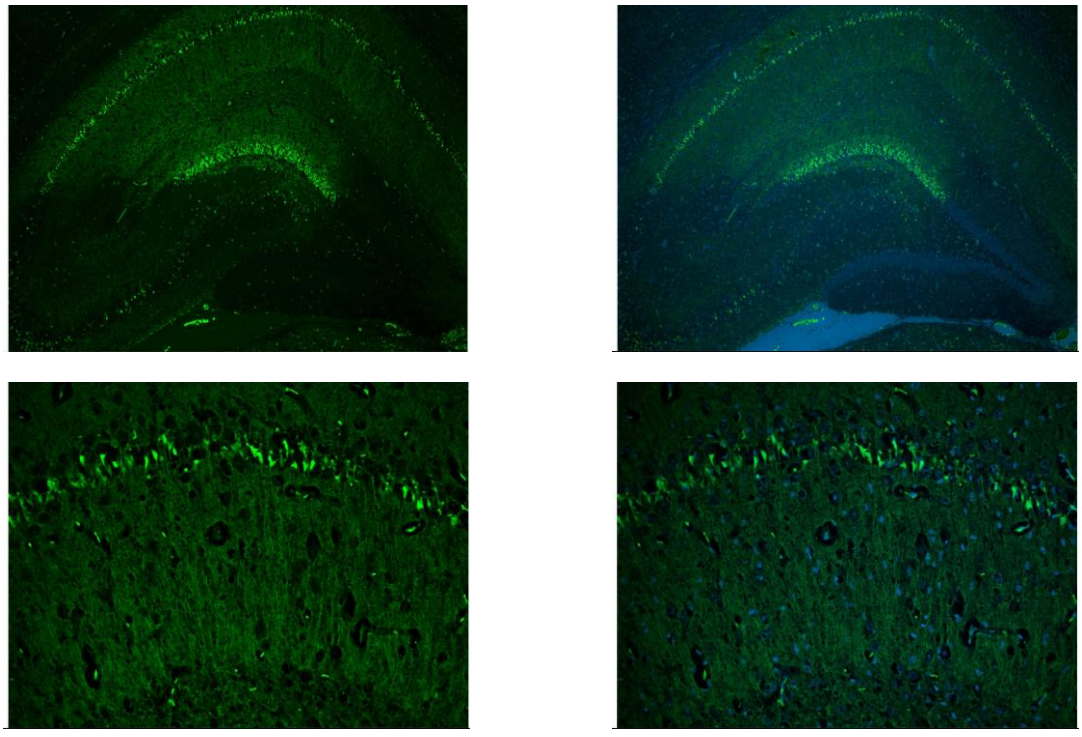
SIze: 5 mL, 50 mL

Spectral Properties: Ex/Em maxima: 497/520 nm

Fluorojade C Stain is an anionic green fluorescent dye. These dyes stain degenerating neurons and their processes after exposure to a variety of neurotoxic insults in brain sections and cultured neurons. The mechanism of neuronal staining by anionic fluorescent dyes has not been determined. It has been proposed that the negatively charged dyes bind to positively charged polyamines or other molecules specifically generated in dying neurons.

Storage & Handling: Shipment at room Temperature. Upon Receipt store at -20° C and protect from light.

Hippocampal degenerating cells are marked with FluoroJade C (green); nuclei marked by DAPI (blue) 72 hours after pilocarpine–induced status epilepticus in rat.



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References**:** Schmued, L.C. and Hopkins, K.J. Fluoro-Jade: Novel Fluorochromes for Detecting Toxicant-Induced Neuronal Degeneration. Toxicol Pathol 28, 91 (2000). Schmued, L., Stowers, C., Scallet, A., and Xu, L., Fluoro-Jade C results in ultra high resolution and contrast labeling of degenerating neurons. Brain Res., 1035, (2005) 24-31.

Staining protocol for tissue sections

1. Mount vibratome sections on gelatin coated slides and dry on a slide warmer at 50- 60^{o}C for at least 30 minutes. For frozen sections, warm slides to room temperature. For paraffin sections, deparaffinize, rehydrate to water, and proceed to step 5.

2. Fix sections in basic ethanol (100% ethanol/0.2% NaOH) for 5 minutes at room temperature.

3. Incubate slides in 70% ethanol for 2 minutes.

4. Incubate slides in dH_2O for 2 minutes.

5. Incubate slides in 0.06% potassium permanganate in dH_2O for 10 minutes.

   Note: potassium permanganate reduces background fluorescence, but may alter protein antigens in tissue sections; the potassium permanganate incubation time may need to be reduced for combined Fluorojade C/ immunofluorescence staining.

6. Rinse slides twice with dH_2O, and incubate in dH_2O for 2 minutes.

7. Prepare 1X Fluorojade C staining solution by diluting 1000X Fluorojade C stock solution 1:1000 in 0.1% acetic acid in dH_2O.

   Note: Use 1X Fluorojade C staining solution within one day.

   Optional: for blue fluorescent nuclear counterstaining, add DAPI to 1X Fluorojade C staining solution at a final concentration of 1 ug/mL.

8. Incubate slides in 1X Fluorojade C staining solution for 10 minutes.

9. Rinse slides 3 X 1 minute in dH_2O.

10. Air dry slides on a slide warmer at 50-60^{o}C for at least 5 minutes.

11. Incubate slides in xylene for 1-5 minutes.

12. Coverslip slides with DPX mounting medium.

13. Image fluorescence using a fluorescein filter set