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Assessment of autophagic flux assay in the adult Drosophila brain

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We use this protocol and it's working

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Abstract

Transgenic flies expressing GFP-mCherry-Atg8a (obtained from the Bloomington *Drosophila* Stock Center) under the control of the neuronal nSyb-GAL4 driver are used for assaying autophagic flux.



- 1 Transgenic flies expressing GFP-mCherry-Atg8a (obtained from the Bloomington *Drosophila* Stock Center) under the control of the neuronal nSyb-GAL4 driver are used for assaying autophagic flux
- 2 Brains from 10-day-old animals are dissected in ice-cold 1X PBS
- 3 The dissected brain are transferred to a glass slide
- 4 The remaining 1X PBS is removed from the slide
- 5 A drop of DAPI Fluoromount is added and the brain mounted with a coverslip
- 6 The dissected brain is imaged immediately within 5 minutes using a Zeiss confocal microscope under a 63X objective lens, and a line scan performed
- 7 For quantification, the intensity of GFP and mCherry is measured per puncta using ImageJ, and the ratio between GFP and mCherry calculated