A simple method for long-term vital-staining of ciliated epidermal cells in aquatic larvae

Supplementary Information

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Figure S1. Young larvae from control experiments (A, only DMSO incubation; B and C, DiI without Pluronic F-127) and DiO stained larva (D), cLSM (magenta: DiI, cyan: DiO).

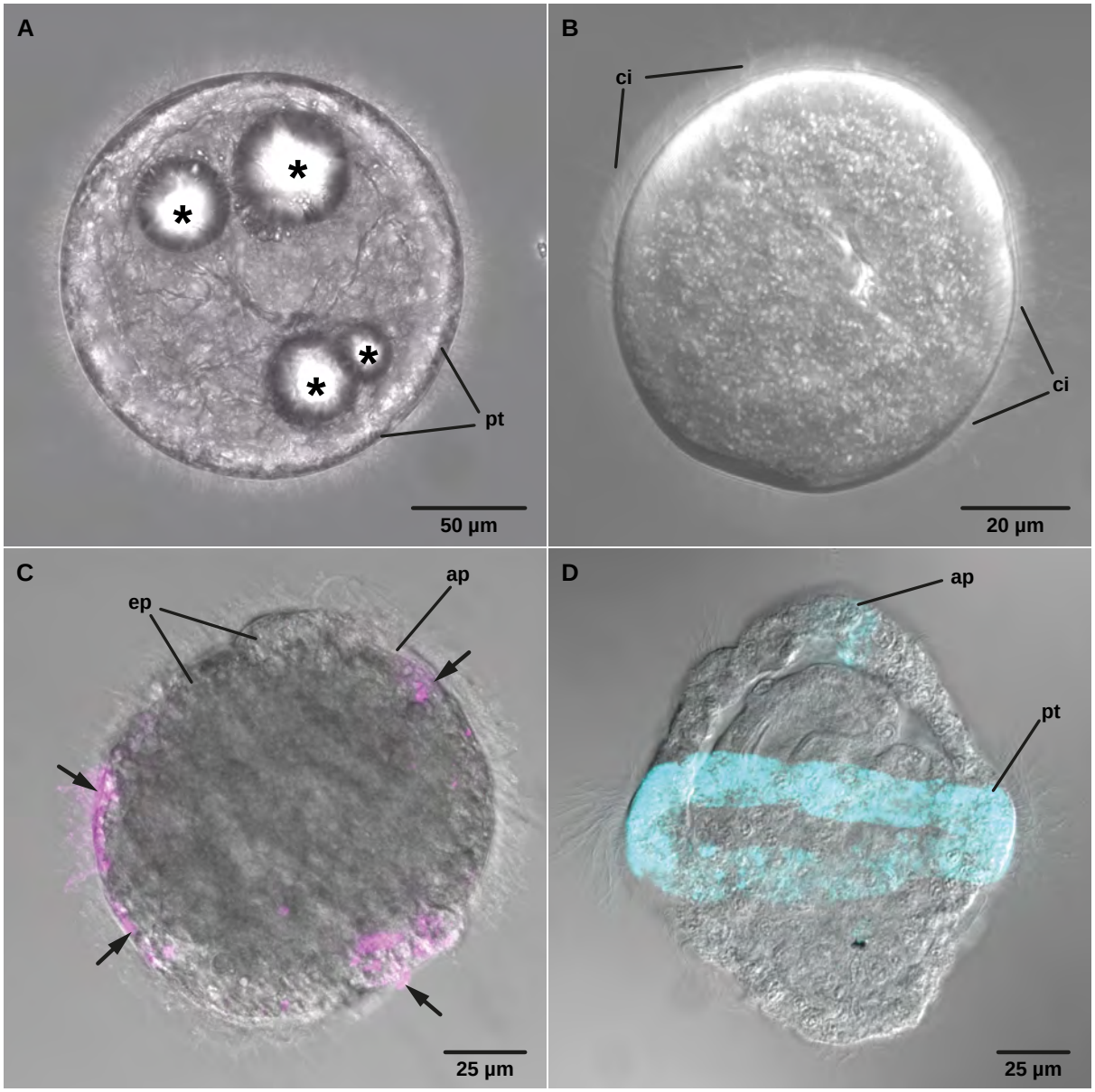

Figure S1: Young larvae from control experiments (A, only DMSO incubation; B & C, DiI without Pluronic F-127) and DiO stained larva (D), cLSM (magenta: DiI, cyan: DiO). A. *Platynereis dumerilii*, 24-hours old larva, maximum projection (apical view): the ciliated prototroch cells (pt) are unstained. Note the large yolk vesicles in the four gastrulated macromeres (asterisks). **B.** *Mytilus edulis*, 21-hours old larva, maximum projection of 32 upper-most sections (28.2 µm; anterior to upper right): epidermal cells with cilia (ci) show no DiI signal. **C.** *Cephalothrix oestrymnica*, 2-day old larva, maximum projection of 5 middle sections (4.4 µm; anterior to upper right): only patchy DiI signals (arrows), due to contact with precipitated DiI crystals during incubation, are visible in some epidermal cells. The majority of the epidermis (ep) including the apical plate (ap) are unstained. **D.** *Thalassema thalassema*, 44-hours old larva stained with DiO/Pluronic F-127 (1µg/ml DiO, 0.02% (w/v) Pluronic F-127, 0.2% (v/v) DMF, and 0.2% (v/v) DMSO in sea-water for one hour at 18° C, fixed for 30 min in 4% FA in 0.1M PBS with 3.42 mM EDTA), maximum projection whole stack (fluorescence excited with λ_{ex} = 488 nm) combined with 5 middle sections (transmitted light, 4.4 µm; anterior is up): fluorescent signals detectable in the ciliated prototroch (pt) and in some ciliated cells of the apical plate (ap).