

Wing Disc Fixation and Immunostaining

Ed Eivers 2008

- Dissect wing discs out of third instar larva in cold 0.02% Triton X-100 PBS (PBST) solution
- Fix discs in 4% formaldehyde for 10 minutes on ice and rinse using PBST
- Incubate discs in blocking solution (2.5% BSA and 5% goat serum in PBS/0.02% Triton X-100) for 1-2 hours at room temperature
- Incubate primary antibodies in blocking solution overnight at 4°C
- Wash 10x for 2 hours in PBST
- Incubate discs for 1-2 hours in blocking solution
- Incubate for 1 hour in secondary antibody at room temperature
- Place wing discs in DAPI-containing Vectashield (Vector) overnight and mount on glass slides