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Western Blot Sample Prep- Whole Drosophila Embryos

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Sonia M Hall

University of Massachusetts Medical School

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Before start

It is important to plan the timing of the embryo collection in advance. To select a specific genotype, be sure to cross in the selectable balancer that will allow you to distinguish mutants from non-mutants. To prevent the collection of unfertilized animals into sample, it is helpful to select animals that have developed far enough through embryogenesis that auto-fluorescence of the gut can be observed.

Example: To collect stage 17 Mcr[P1] animals: Setup a four hour collection and age for 17 hours at 25 degrees Celsius. For homozygous mutants, collect non-YFP animals and select only animals that have auto-fluorescence in the gut.



- 1 Collect 100 embryos and place into eppendorf tube (1.5ml) containing 50ul of embryo wash

 50 μ L

- 2 Add 3 drops of bleach and let sit for 3 mins.

 150 μ L

 00:03:00

- 3 Raise volume to 1.5ml using dH₂O
Spin down for 1min

 1500 μ L

 00:01:00

- 4 Remove solution
Add 1.5ml of dH₂O
Spin down 1min

 00:01:00

- 5 Remove solution
Add 1.5ml of dH₂O
Spin down 1 min

 1500 μ L

 00:01:00

- 6 Remove solution with micropipette
Add 50ul of 1X PBS

 50 μ L

- 7 Add 50ul of 2X SDS SB

 50 μ L

- 8 Homogenize with pestle and 27ga needle



9 Boil for 5 mins at 98 degrees C

 00:05:00

10 Store at -20 degrees C