



AQUA-FAANG – Standard Operating Protocol Cross-linking DNA ready for ChIP-Seq using salmonid embryos

Overview: This protocol describes a method used to crosslink proteins to DNA of whole salmonid embryos.

Consumables:

- 1% PFA solution
- Rotator system
- 1M glycine solution
- PBS
- Fixed angle centrifuge

Repeat each step for all biological replicates.

1. Pre-chill fixed angle centrifuge
2. Starting from step 2.11 of DevMap SOP - Embryo extraction protocol. Remove supernatant and resuspend in 875ul of 1% PFA.
3. Put tubes on a rotator at 50rpm for X min, at room temperature.

Note: X min = 8 mins for blastula and gastrula, 10 mins during segmentation stages, 15 mins post-segmentation stages

4. Add 125ul of 1M glycine. Continue rotating for an additional 10 mins at 50rpm, at room temperature.
5. Centrifuge at 300RCF, for 6 min at 4°C.
6. Discard supernatant and resuspend in PBS.
7. Store at -80°C