Frozen sample processing for mouse embryonic heads

1. Dissect mouse embryonic heads in 1XPBS.
2. Fix the samples in 10% Formalin O/N at RT or 4% PFA O/N at 4°C.
3. Wash the samples in 1XPBS.
    a. For samples younger the E14.5, proceed immediately to step 4 for dehydration.
    b. For samples older than the E14.5, decalcifiy in 10% EDTA at 4 °C first then proceed to step 4 for dehydration. The decalcification time is dependent on the stage of the embryos.
4. Dehydrate the samples in 15% sucrose/PBS O/N at 4 °C.
5. Dehydrate the samples in 30% sucrose/50% OCT
    a) For E10.5 samples, dehydrate in in 30% sucrose/50% OCT for 1 hr at RT
    b) For E11.5 samples, dehydrate in in 30% sucrose/50% OCT for 2 hrs at RT
    c) For E12.5 samples, dehydrate in in 30% sucrose/50% OCT for 3 hrs at RT
    d) For E13.5 or older samples, dehydrate in in 30% sucrose/50% OCT overnight at 4 °C.
6. Embed the samples in OCT on a dry ice block.
7. Store the samples at -80 °C.