Xgal staining

This protocol assumes you already have fixed, hydrated and washed tissue. Heat fixation will not work for this protocol: use aldehyde fixation only. Tissues should not be treated with Methanol before Xgal staining, because Methanol inactivates the enzymatic activity of the β -Galactosidase enzyme.

- 1. Prepare Xgal staining buffer
- 2. Incubate tissue in staining buffer at 37°C (water bath or dry incubator are both fine) until you see blue staining. This may take anywhere from 15 minutes to overnight.

Xgal staining buffer: all you need to do is mix these things together – make up FRESH solution every time and take care of your stock solutions in the meantime

	original protocol volume	original protocol stock concentrations	our stock concentrations (if different from original)	our protocol volumes
	62 µl	50 mM potassium ferricyanide		62
	62 μl	50 mM potassium ferrocyanide		62
	15 μl	20% Triton X-100	10%	30
	30 µl	5M NaCl		30
	1 μl	1M MgCl2	50mM	20
	100 μl	10x PBS		100
	697 μl	water		675
	33 µl	8% Xgal in DMF (add this last)	50mg/ml	21
FINAL VOLUME	1000 ul			1000 ul