

# Combined LacZ staining and *in situ* hybridization

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## Reagents

[In vitro transcription of riboprobe](#)

[Embryo fixation and permeabilization](#)

[Hybridization and post-hybridization washes](#)

[Immunodetection and development of signal](#)

[LacZ staining](#)

## Method

- 1 Dissect out the embryos in cold PBS.
- 2 Incubate the embryos in cold LacZ fixative ([LacZ staining](#)) for 10 min at 4 °C.
- 3 Wash the embryos in cold PBS plus 0.02% NP40 ([LacZ staining](#)) for 10 min at 4 °C.
- 4 Stain the embryos for the minimal period of time in LacZ staining solution ([LacZ staining](#)) at 37 °C until sufficient colour has developed.
- 5 Wash the embryos three times for 20 min in PBS plus 0.02% NP40 at room temperature.

Process the embryos for non-radioactive whole-mount *in situ* hybridization according to:

[In vitro transcription of riboprobe](#)

[Embryo fixation and permeabilization](#)

[Hybridization and post-hybridization washes](#)

[Immunodetection and development of signal](#)