

Thaw Embryos in Vials

Reagents, supplies, and equipment:

- M2 medium (Millipore MR-015P-5D)
- KSOM medium (Millipore MR-020P-5D)
- 35 mm culture dishes
- PBS (Lonza 17-512F)
- Selectapette (Clay Adams)
- Mineral Oil (Sigma M5310)

Keywords: [vials](#) [8-cell](#) [cryopreserved](#) [embryos](#) [cryo vials](#)

Expand

Thawing 8-cell stage embryos in vials (frozen using JAX slow freeze method):

1. Remove cryo vial from LN2 storage and place on the bench at room temperature until thawed, about 12 to 15 minutes.
2. When completely thawed, slowly add 0.8 ml of PBS to the cryo vial in a dropwise fashion to dilute the DMSO.
3. The contents of the cryo tube are withdrawn by a Selectapette or wide bore pipette tip.
4. Wash the embryos in a fresh drop of M2. Wash embryos through 8 to 10 drops of media. Place embryos in drops of KSOM overlaid with mineral oil.
5. Transfer the embryos either into the oviducts of a 0.5 dpc pseudopregnant mouse. Or you may choose to culture the embryos to the blastocyst stage in KSOM and transfer to the uterus of a 3 dpc recipient.