COSMO BIO CO., LTD.

mR1ECM
(*in vitro* culture of rat embryos)

Cat. No. CSR-R-M174

CSR-R-M191

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*Keep them in 4℃ until use. Use all the media once opened and avoid using the remaining because of the quality degradation.

Superovulation induction and mating

Mature female Wistar rats (8-12 weeks old) received intraperitoneal administration of PMSG (150 IU/kg/rat) and hCG (75 IU/kg/rat) at 48-hour intervals to induce superovulation. After hCG administration, they were mated with male rats. On the next day, vaginal plugs were confirmed to ensure mating. Mated female rats were used in the experiment. Pronuclear-stage embryos can be collected on the day of vaginal plug confirmation, and 2-cell-stage embryos on the next day.

Recommended schedule

PMSG: 11 : 00 -13 : 00
hCG : 11 : 00 -13 : 00 (48 hours after injecting PMSG)
Collection time
Pronuclear-stage fertilized oocytes : 15 : 00-16 : 00 (on the day of vaginal plug confirmation)
2-cell-stage embryos 15 : 00-16 : 00 (on the next day of vaginal plug confirmation)

Preparation of drops

Place 3 drops of mR1ECM (100 µL each) into a dish. Cover them with liquid paraffin and incubate (5% CO_2) for at least 30 minutes for gas equilibrium.

Collection of embryos (oviduct flushing)

Disinfect all dissectors with alcohol.
Heat culture medium for flushing (M2, Cat No. CSR-R-M083 and CSR-R-M084 or PB1, Cat No. CSR-R-P138 and CSR-R-P138) to 37℃ .

1. Euthanize a mature female rat. Pull out the uterus, ovary, and part of fat using scissors and forceps. Cut only the oviduct on a filter paper, and remove blood.
2. Insert glass capillary or flush needle to fimbria of the collected oviduct, and flush the culture medium.
   * If you collect Pronuclear-stagefertilized oocytes, please add hyaluronidase (final concentration: 0.1%) to the culture medium for flushing(M2 or PB1).
3. Transfer the embryos into the mR1ECM drops.
4. The resulting two-cell stage embryos can be cultured *in vitro* to blastocyst stage. For culturing, use mR1ECM (400 µL drop).

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