COSMO BIO CO., LTD.

0.25M sucrose
(for fusion of frozen oocyte / embryo)

Cat. No.	CSR-R-Y077	Size:	10 *2 ml
	CSR-R-Y078		10 *5 ml

www.cosmobio.com

*Store at 4℃ until use. As a quality degrades after opening, use up all contents at one time.

Preparations

1. 0.25M sucrose is warmed to 37 ℃ before use.
   *In case warming with CO_2 incubator, put in the ample, gas equilibration should not be done because 0.25M sucrose solution reacts with CO_2 and pH level may change. (Put in the incubator without breaking neck of ampoule.)
2. Prepare three drop balls of 100µl KSOM (or mWM) on the dish according to the number of tube for fusion. Drops are left to stand in incubator for more than 30 minutes and are applied after gas equilibration.

Process of fusion

1. Take a cryotube out of liquid nitrogen tank and remove a cap promptly. Discard remaining liquid nitrogen in a tube and stay at room temperature for 30 seconds.
2. Add 0.9mL of 0.25M sucrose solution warmed at 37℃ in advance with micropipette to the tube and transfer to dish by quick pipetting.
   *As the stock solution after fusion has strong cytotoxicity, add 0.25M sucrose into a tube and pipette quickly until the stock solution is completely fused.
   This operation is carried out as promptly as possible. This process is very important to maintain good viability of embryo.
3. Cowash a tube with 0.4-0.5mL of 0.25M sucrose.
4. Collect embryos from liquid mixture of (2) & (3), transfer to a drop of KSOM (or mWM) and stay for 10 minutes.
5. Wash surviving embryos twice with KSOM (or mWM) drop (move a drop from same dish).
   *The 2-cell stage embryos obtained from the process are just possible to apply to in vitro culture in the drop of KSOM (or mWM) without changing culture medium.

COSMO BIO CO., LTD.
TOYO EKIMAE BLDG. 2-20, TOYO 2-CHOME,
KOTO-KU. TOKYO 135-0016, JAPAN
TEL : (81)3-5632-9617
FAX : (81)3-5632-9618
e-mail : export@cosmobio.co.jp
URL : www.cosmobio.com