COLLECTION AND CULTURE OF 1-CELL EMBRYOS

Required Materials:

MEM-HEPES

Hyaluronidase

(3) 35 mm Petri dishes

1.7 ml microcentrifuge tubes

Forceps and dissection scissors

Before beginning:

1. The day before collection, superovulate 7-week-old female mice according to protocol (see: *Superovulation* protocol).

2. Add 2 ml MEM-HEPES to each of three (3) 35 mm Petri dishes for collection and preheat medium to 37^{o}C for at least 30 minutes.

Collection of 1-cell embryos:

1. Sacrifice superovulated female mice and dissect out the oviducts. Place oviducts in prewarmed MEM-HEPES in the first collection dish.

2. Examine oviducts under a microscope. The section of the oviduct containing the 1-cell embryos will appear swollen. Isolate the swollen sections of the oviducts and transfer them to the second collection dish.

3. Add 20 µl hyaluronidase to the dish to remove the cumulus cells from the embryos. Incubate on the warming plate for about 10 minutes until the cumulus cells have fallen away from the embryos. Transfer cumulus-free embryos to third collection dish.

4. Culture embryos in the appropriate medium (eg, KSOM supplemented with essential and non-essential amino acids).