

## Preparation And Shipment Of Biopsy Sample For PGT Day 5 Trophectoderm Biopsy

- 1. Embryos destined for trophectoderm biopsy should be artificially hatched sometime on Day 3 post-retrieval. The hatching helps trophectoderm cells to herniate through the zona pellucida by Day 5 (D5).
- 2. On D<sub>5</sub>, observe the embryos at a convenient time (around mid-morning) to determine which embryos have herniating trophectoderm that can be biopsied.
- 3. Perform the biopsy as late as possible on D5 (to allow as many embryos as possible to herniate), but keep in mind that the biopsy must be finished in time for the samples to be packed and shipped overnight to the PGD Lab.
- 4. The embryo biopsy may be performed in whichever medium your laboratory normally uses. Try to obtain a sample of approximately 4-6 trophectoderm cells. We recommend changing biopsy tools after each embryo is biopsied. After the biopsy, keep the embryos separated by moving them to individually numbered droplets in D5 culture medium. Freeze or vitrify the biopsied embryos after the biopsy is complete.
- 5. Prepare two rinse droplets of approximately 20 uL each for each biopsy sample using the provided phosphate buffered saline with fetal bovine serum (PBS/FBS). Rinse the biopsy sample through each droplet, then transfer the sample, in approximately 2.5 uL of PBS/FBS, to one of the provided 0.2 mL thin-walled PCR tubes. Use a new pipet tip for each sample. Each PCR tube should be clearly labeled using a permanent marker with the patient's initials and the embryo number. Using a mini centrifuge (i.e. picofuge), pulse spin the tube containing the trophectoderm biopsy for a few seconds to ensure the cells remain in the buffer at the bottom of the tube at the time of freezing. Place each sample in a -20°C or -80°C freezer immediately.
- 6. Prepare a negative control sample by placing 2.5 uL of the provided PBS/FBS into another labelled 0.2 mL thin walled PCR tube. We require one tube of a negative control per patient per day of the biopsy. Place the negative control sample in the freezer with the biopsy samples. The PGD Lab will prepare an appropriate positive control sample for the diagnostic assay.
- 7. Place all of the biopsy samples and the negative control sample in the provided tube rack with the lid securely closed and freeze at -2oC until shipped. Ship samples to the PGD Lab using the provided shipping instructions. Please complete the provided "Biopsy Record" and send it with the biopsy samples. In addition, please send an e-mail to the PGD Lab provide us with a tracking number for the shipment.