WHAT TO EXPECT WHILE YOUR EMBRYOS ARE IN THE IVF LABORATORY

The laboratory component of your IVF journey should not be mysterious, and we always welcome your questions. The science behind growing embryos in vitro is constantly evolving as we strive to make the culture environment as natural as possible to optimize your chance of success.

Recent studies have shown us that growing embryos for 5 or 6 days before transfer and freezing (when they will have reached the “blastocyst” stage) has some crucial advantages:

1) Growing embryos for 5 days instead of 3 is an excellent selection tool: only a proportion of embryos will become blastocysts, the others will stop growing.

2) If the embryo becomes a blastocyst it has a much higher chance of implanting. This means fewer embryos can be transferred in order to achieve a healthy pregnancy, and the risk of multiples is subsequently reduced. It also allows us to eliminate poorer quality embryos which would not have made a pregnancy.

3) For successful implantation, the stage of the embryo and the lining of the uterus must be “in sync”, and this is more easily achieved when the embryo is at the blastocyst stage, matching more closely with what happens in a natural cycle.

At PCRM we have a reliable, reproducible culture system, and because of this we are happy to grow everyone’s embryos to day 5 before transfer. There is no way to know for sure whether all embryos that fail in culture would also have failed in utero, but we believe that there is no harm caused to your embryos by five days of growing in the lab. What follows is a brief outline of what will be happening to your embryos for the six days that they will be in the Lab.
(Day ZERO is the day of your egg retrieval).

Normally fertilized embryos show 2 pronuclei (PNs) in their centre on the morning of day 1 (one from the male and one from the female). They will not divide until later today. We usually call with your fertilization report between 8 and 10am.

On day 2 we expect that most of the embryos will now have divided, and will ideally be 2-4 cells. Around 98% of embryos which have shown normal fertilization on d1 will divide and continue to grow.

(No phone call)

On day 3 we expect the embryos to have made another division from the previous day, and to ideally have 6-8 cells. Up until day 3, it is only the maternal DNA which has been contributing to embryo development. At some time on day 3, the male contribution kicks in, so there is sometimes a change in embryo quality after d3, especially if there is a known “male factor”.

(No phone call)

On day 4 we expect the embryos to have more than 8-10 cells, and to have started to compact – this is the morula stage (this just means that individual cells can no longer be counted, and that the cells have started to fuse together). You will be called with an update and your transfer time.

On day 5 we expect a proportion of the embryos to have become blastocysts. The grades and quality of your embryos will be fully explained to you at the time of the transfer. Any surplus embryos which form GOOD QUALITY blastocysts on d5 or d6 may be cryopreserved for future use. Read the explanation of our blastocyst scoring system later in this booklet.
WHEN WILL YOU CALL ME WITH A DAILY UPDATE?
- We will normally try to call you between 8 and 10am with fertilization results and embryo updates. However, do not worry if our call is a little later (especially on weekends): it just means we are busy in the Lab, but we will not forget to call! We will NOT call you on d2 or d3, as we prefer to leave the embryos completely undisturbed.

DO YOU GRADE THE EMBRYOS?
- From day 1 to day 4, we score embryos on a scale from 1 - 5. 1 = excellent  2 = good 3 = average 4 = a little below average 5 = below average (poor). If embryos are graded 1-3, we are happy with their quality. If they are graded 4 and 5, we will caution you that those will likely have a lower chance of success. The most important thing is that there is daily progression of the embryos. It is also perfectly normal to have a mixture of grades, especially when you have a larger number of embryos.

- Once the embryo becomes a blastocyst on day 5, its score describes the blastocyst, and consists of a number followed by 2 letters (for example 4AA). This scoring system and what it means will be fully explained to you before your transfer. Like most IVF Clinics, we use the Gardner grading system for blastocysts.

EMBRYO TRANSFER – IMPORTANT INSTRUCTIONS
- You will be given your transfer time when we call with your update on day 4. Transfer time is usually mid to late morning. Please remember to arrive at PCRM 30 minutes before the scheduled time. Take your medications as you normally would on the morning of your transfer, and make sure that your bladder is comfortably full - this will make it easier for the Physician to have a clear picture for your ultrasound-guided transfer. Drink 3-4 glasses of water 1-2 hours before the scheduled transfer time.

WILL YOU FREEZE MY SURPLUS EMBRYOS?
- On average, we expect 40-50% of good quality day 3 embryos to develop into blastocysts. That means that freezing of surplus good quality embryos is done for 40-50% of patients. Blastocysts have to be of good quality or they will not survive the freezing or thawing processes. We will not freeze blastocysts of poor or sub-optimal quality, as they are not likely to survive.

WHY DO YOU CULTURE THE EMBRYOS FOR 5 DAYS?
- Blastocyst culture is used as a selection tool, since we know that only a proportion of embryos will develop to the blastocyst stage. We are confident in our lab culture system and although we cannot know for sure, we believe that what happens during their 5 days of growth in the lab is what would happen inside of you. That is why we are happy to transfer on day 5 for all patients.

- Sometimes your embryos will not have reached the blastocyst stage at the time of your transfer on day 5 – do not worry, this happens fairly often. As long as your embryos have continued to grow each day, we will still transfer the one or two most advanced embryos, and grow all of the others for one more day. You will receive a final call from the Lab on d6 to let you know if any embryos were frozen.
EMBRYO GRADING FOR CLEAVAGE STAGE EMBRYOS (day 2 and 3)

**Excellent quality**
- No irregularities of membranes and/or cytoplasm
- Blastomeres of even shape and size
- No fragmentation
- No granularity of cytoplasm

**Good quality**
- Minor irregularities of membranes and/or cytoplasm
- Blastomeres of slightly uneven size
- ≤10% fragmentation

**Average quality**
- Multiple minor irregularities OR a single moderate irregularity
- >10 and up to 25% fragmentation

**Below average quality**
- Granular cytoplasm, dark colour, irregular shape/membranes/zona
- >25 and up to 50% fragmentation

**Poor quality**
- Multiple abnormalities of zona, membranes and/or cytoplasm.
- >50% fragmentation.
• If your embryos are graded 1, 2 or 3 we are happy with their quality, and these are the grades we see for most people each day in the lab.
• Most patients will have a mixture in the quality of their embryos and this is normal, especially if you have a larger number of embryos.
• If your embryos are graded 4 or 5, they are of poorer quality. This does not mean they will not lead to a pregnancy, but we may caution you that they are less likely to make blastocysts, and less likely to implant.
• Remember, even if all of your embryos are perfect on day 3, on average only 40-50% of them will become blastocyst on day 5.
BLASTOCYST GRADING SYSTEM – DAY 5 AND DAY 6

Inner cell mass (ICM) Grading:
A. Tightly packed, many cells
B. Loosely grouped, several cells
C. Very few cells

Trophectoderm Grading:
A. Many cells forming a cohesive epithelium
B. Few cells forming a loose epithelium
C. Very few cells
On average, 40-50% of embryos which are “good” quality on day 3 will progress to blastocyst stage.

The number in the score refers to the size of the blastocyst, and it changes quite rapidly on day 5.

Inner Cell Mass (the first letter in the score) = the tight ball of cells inside the blastocyst which will eventually become fetal tissue.

Trophectoderm (the second letter in the score) = the surrounding cells which will become the placenta.

Grades A and B, expanded to size 2, 3 and 4 are all good quality embryos and can be frozen. There is little difference in the potential to implant between blastocysts graded AA or BB.

Blastocysts with a “C” grade have a lower chance of implanting but do still make pregnancies. Grade Cs are not usually frozen as they are considered poorer quality, and unlikely to survive the freeze/thaw procedure.