START: Implantation and early pregnancy
From: Stem Cell Information, NIH USA 2006
http://stemcells.nih.gov/info/scireport/appendixa

Day 0:
- Fertilization
- Ovulation
- Oocyte

Day 1:
- Fertilized egg (zygote)

Day 2:
- First cleavage
- 2-cell stage
- 4-cell stage

Day 3-4:
- 8-cell uncompact morula
- 8-cell compact morula

Day 4:
- Early blastocyst
- Blastocoele
- Inner cell mass

Day 5:
- Trophoderm
- Early blastocyst

Day 6-7:
- Late-stage blastocyst (hatching)
- Zona pellucida

Day 8-9:
- Implantation of the blastocyst
- Epiblast
- Hypoblast
Implantation: The Science

- The early embryo enters the uterine cavity as an 8 cell morula and becomes a 30-200 cell blastocyst before implantation.
Implantation : The Science

• The process of implantation begins with apposition and adhesion of the blastocyst to the uterine epithelium.

• This occurs 2-4 days after the morula enters the uterine cavity.

• The process is mediated by cytokines and involves adhesion molecules (integrins) that interact with the extracellular components eg. Laminin and fibronectin.
Implantation: The science

- The embryo hatches from the zona pellucida 1-3 days after the morula enters the endometrial cavity.

- The endometrium is prepared for implantation by the complex activity of cytokines, growth factors and lipids.

- These factors are modulated by the sex hormones (mainly progesterone)

- *The endometrium has a receptivity window of only a few days.*
Implantation

- **Hatching** leaves the blastocyst free of the zona pellucida.

- It should have occurred approximately at the end of the uterine tube or in the body of the uterus.

- The embryo floats in the uterine glands rich mucus secretion and able to directly access this nutrition for continued growth.
Implantation

-The blastocyst initially weakly adheres to the endometrial wall rolling across its surface.

-Increased adhesion may lead to attachment, **adplantation**, on the inner cell mass side of the blastocyst

-This will be the site where **implantation** will begin and the placenta will develop.
Implantation

-Trophoblast cells at the site of adplantation proliferate and form an additional layer, the syncitiotrophoblast layer.

-This layer of cells rapidly divide, secrete enzymes that degrade the endometrial extracellular matrix and secrete human Chorionic Gonadotropin (hCG).
Implanting blastocyst
Implantation: The Science

• Trophoblastic invasion rapidly follow adhesion of the blastocyst.

• This is mediated by proteinase degradation of the extracellular matrix.

• The placenta is formed in the second week after ovulation.
Implantation Detection

• **When is a urinary bhcg first positive?**
  – 3 days before the period is due
  – Timing (early morning or evening etc.) affects result
  – Sensitive to about 25 units/litres using a monoclonal antibody to the beta subunit of HCG.

• **When is the blood test bhcg positive?**
  – 8 days after ovulation
  – 1 day after implantation
  – A positive test by the time of the expected period will be 98% correct.

• **When is the trigger hcg out of the patients system?**
  – By day 10 post injection.
Implantation Detection

• *Which day of the cycle is the first blood test usually done on?*
  – Day 11 after blastocyst (Day 5) transfer.

• *What is the usual range of the first blood test?*
  – 50-200
Implantation Detection

• *When is the second blood test done, and what is the expected level?*

  – 48 hours after the initial blood test.

  – The bhcg doubles every 48hours in a normally developing pregnancy
Implantation: Medications

*What medications are needed for implantation?*

- Natural thaw cycle
- HRT (FET) Artificial thaw cycle
- IVF cycle
- Egg Donor cycles
Implantation: Frozen Embryo Transfer (FET) cycles

• **Natural cycle:**
  – Patients own Corpus Luteum will provide the ideal amount of progesterone. (9-47ng/ml)
  
  – Some doctors given pregnyl Day 3 and 5.
Implantation: Frozen Embryo Transfer (FET) cycles

• **HRT/ Artificial thaw cycle:**
  – No Corpus Luteum, so patient needs Progesterone support. Crinone, Progesterone pessaries or endometrin
  – Need to continue Estrogen daily (progynova 2mg bd or tds)
  – Continue until 12 weeks of pregnancy
Implantation: IVF cycles

- IVF Cycle:
- 2 common regimes for allowing implantation:

  - **Vaginal Progesterone**
    - Crinone gel (8%) - 1-2 vaginal application daily
    - Progesterone pessaries (200mg) – 3 vaginal tables per day
    - Endometrin 100mg PV TDS

  - **Hcg injections** (Ovidrel, Pregnyl)
    - Pregnyl 1500IU Day 3 and Day 5
    - Risk of OHSS if > 10 follicles
Implantation: Donor egg cycles

• **Fresh Donor egg cycle:**

  -The donor will not need any medications in the post egg collection period.

  -The recipient will generally be on an HRT (artificial cycle) taking progynova 2mg tds and progesterone PV daily starting the day of the donor’s egg collection. This gives flexibility to get the two women’s cycles in phase.
Early Pregnancy: The Science

- Biochemical pregnancy
- Clinical pregnancy
- Ectopic pregnancy
- Blighted ovum
- Miscarriage
Early Pregnancy: The Science

• Biochemical pregnancy
  
  – When the urinary or blood hcg is initially positive, but then it declines and no sac is seen on USS.
  
  – Probably occurs in close to 30% of pregnancies if early bhcg monitoring were done.
  
  – No curette is usually needed, and a period usually follows a few days later.
Early Pregnancy: The Science

• Clinical pregnancy

  – When a sac, a fetal pole and a fetal heart is seen within the uterus.

  – On a scan in an IVF clinic, a sac with a fetal pole and a fetal heart should be seen at 6 and a half weeks after the LMP.

  – The fetal heart rate should be about 140bpm – 60-100 is slow and is a poor prognostic sign.

  – A positive fetal heart at 7 weeks has a 99% chance of an ongoing pregnancy and a 95% chance in the presence of vaginal bleeding.
Clinical pregnancy
Early Pregnancy

• On an ultrasound in an IVF clinic, a sac with a fetal pole and a fetal heart should be seen at 6 and a half weeks after the LMP.

• The hcg should double every 48 hours.
Early Pregnancy: The Science

- **Ectopic pregnancy**
  - When the pregnancy has implanted outside the uterine cavity
  - The hcg level is positive but the uterus is empty at 6 weeks
  - Common if history of Chamydia, ruptured appendicitis or tubal reconstructive surgery.
  - Heterotopic pregnancies increasingly possible with IVF if two embryo’s are replaced. 1 in 300 IVF pregnancies.
  - Recurrence rate of 10%
Ultrasound findings in Ectopic pregnancies

- An *extrauterine GS with a yolk sac* is pathognomonic for an ectopic pregnancy.

- An *empty uterus* on USS with a bhcg of 800-1000 signals a probable ectopic.

- A *pseudogestational sac* “single sac sign” can be seen with an ectopic.

- *Free fluid* in the POD is often seen with ruptured ectopic pregnancies.
Ectopic pregnancy
Early Pregnancy: The Science

- **Blighted ovum**
  - Symptoms of pregnancy still persist.
  - Often needs a D &C.
  - A sac is seen within the uterus, but no fetal pole is seen at >7 weeks.
Early Pregnancy: The Science

• Miscarriage

  – (Complete/inevitable/ missed-abortion/early pregnancy loss)

  – Bleeding is usually noted before pain.

  – May miscarry spontaneously or may require a D&C.

  – 3 consecutive miscarriages constitutes “recurrent miscarriage” and needs to be appropriately investigated.
Early Pregnancy

• **Twin pregnancies:**

• **Dizygotic** – from two eggs.
  – Higher chance in certain races (e.g., Nigerian > Japanese)
  – Higher chance in older women.
  – If replace two embryos and have a positive pregnancy test – 30% chance of a twin pregnancy.

• **Monozygotic** – one egg splits (identical twins)
  – Stable across populations
  – ? Higher in ART but not conclusively shown by studies.
  – Risky pregnancies
Twin pregnancy
Recurrent implantation failure

- Assessment of endometrial cavity
  - Removal of polyps, submucous fibroids

- PGD to exclude aneuploidy
- Donor eggs, embryos
- Surrogacy
Recurrent implantation failure

• Many different non evidenced based treatments,
  – Aspirin, clexane
  – Prednisolone to treat ‘natural killer cells’
  – Acupuncture
  – Viagra
  – Endometrial scratch
Summary:

- Implantation of a pregnancy is still a topic that requires more research.

- There is a lack of evidence supporting many of the adjunctive treatments now used widely in clinical practice.

- Early pregnancy may seem like a “small” biochemical event to the observer, but is “their baby” to many women who achieve this stage of pregnancy only to subsequently miscarry.

- Offer sensible routine advice ..... be sympathetic and supportive.
Thank you