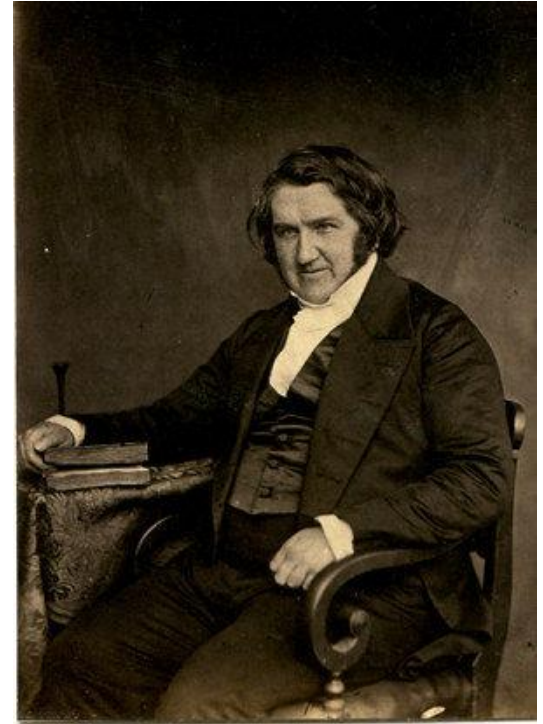


Reproduction in the older woman

William Ledger
University of Sheffield
Centre for Reproductive Medicine and Fertility



Infertility is not a 21st century phenomenon

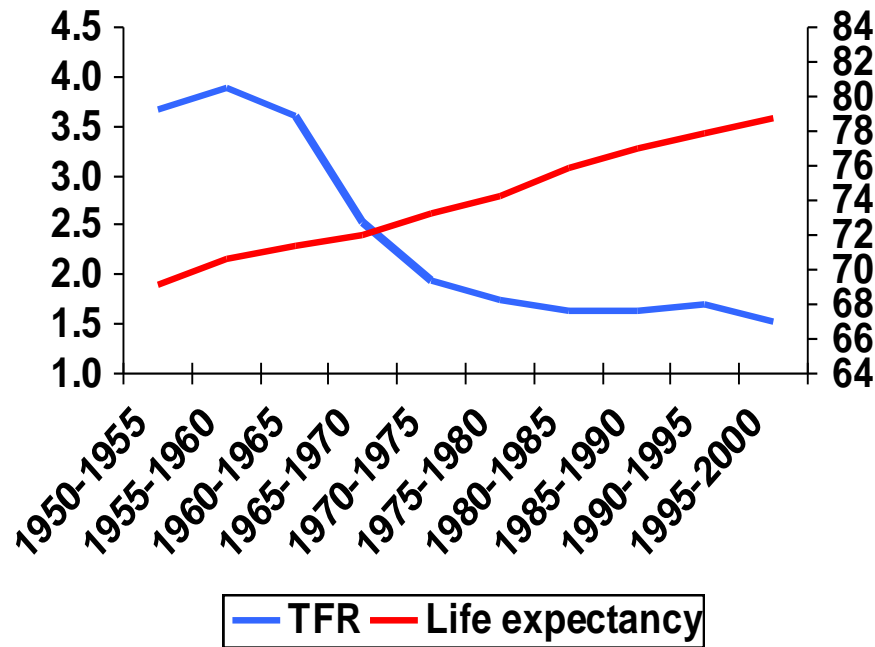


In “marriages which had lasted five years or more, and in which the husbands were under 75 years of age, ... one marriage in 6.5 was unproductive”.

J Y Simpson

Survey of 495 British Peers, cited in Gibbons (1911)

Some things are different in the 21st century



Over the last 50 years life expectancy has increased by
5 hours every day

life expectancy - male - 43.5 in 1900 to 75.4 in 2000

female - 46.0 in 1900 to 81.4 in 2000

By 2050, life expectancy will be 82.3 years for males and 87.4 for females

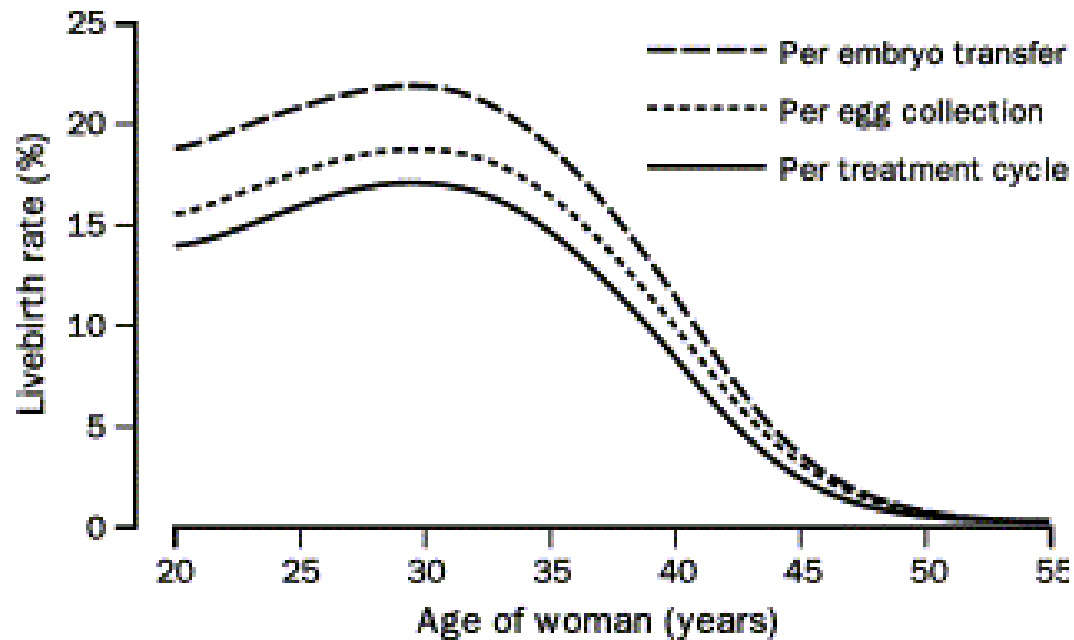
And some things remain the same...

- 2570 North American women
- age 44 - 55
- prospective study for 5 years
- onset of menstrual irregularity at 47.5 years
- duration of transition 4 years
- youngest confirmed livebirth - 5 yrs 3 mths
- oldest - 57 yrs 10 mths

McKinley et al (1992)

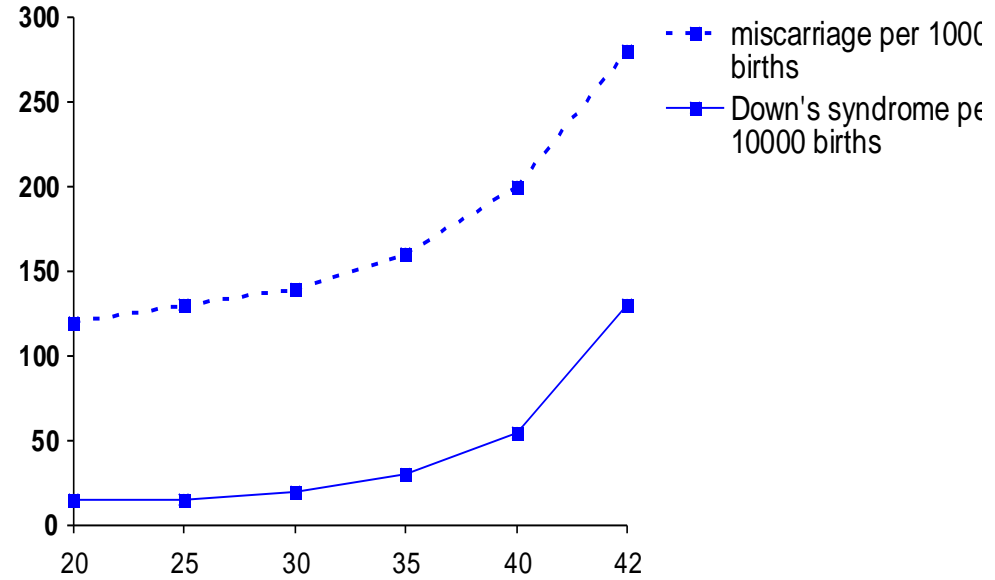
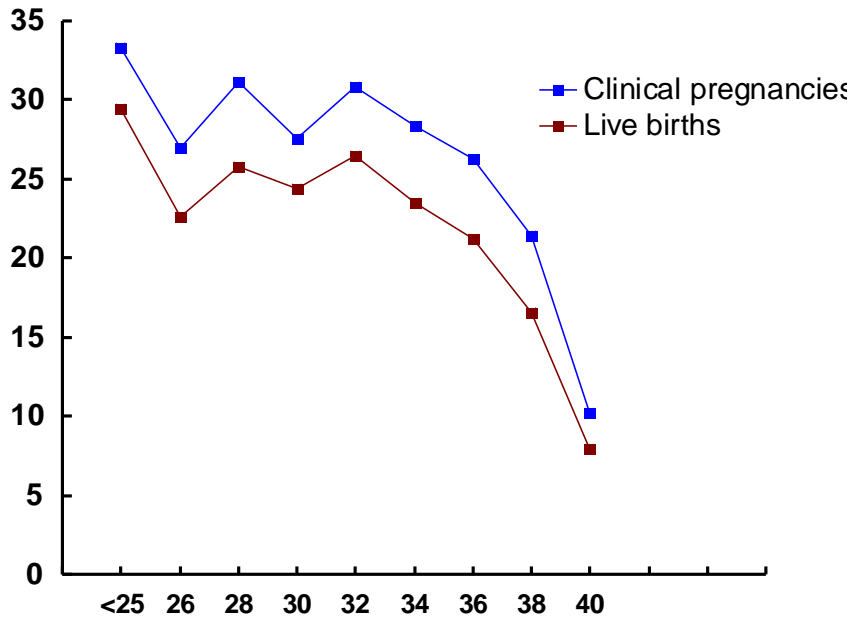
Guinness (2006)

Women do not remain fertile until menopause



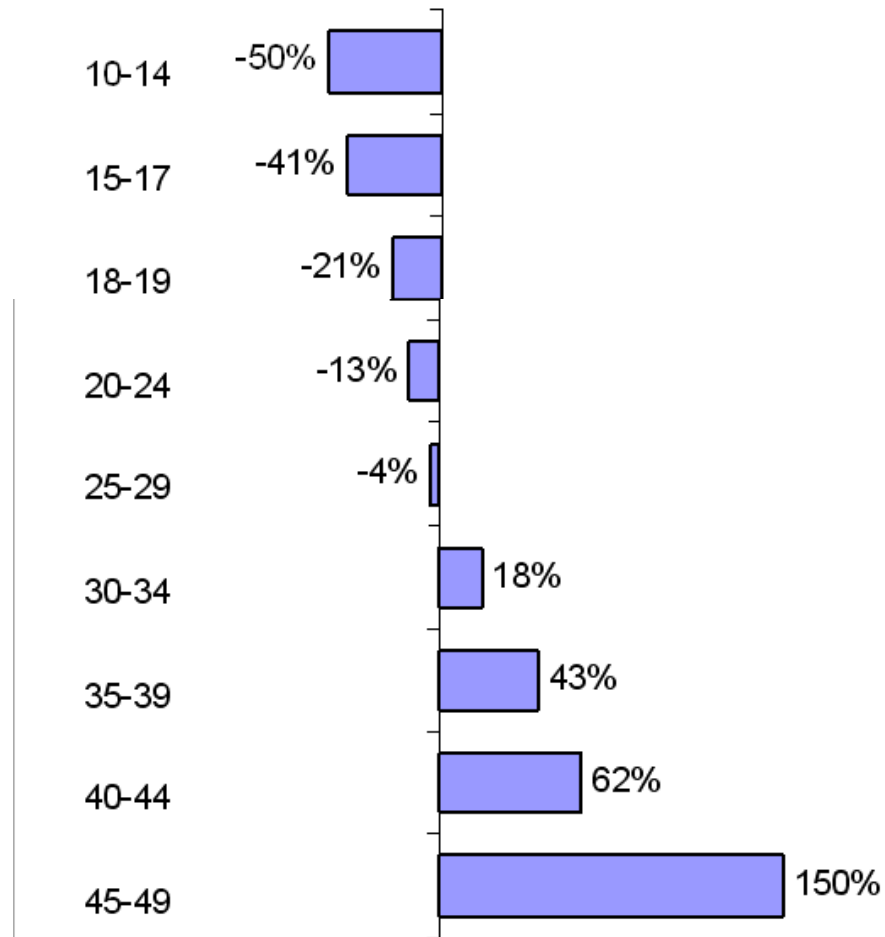
What goes wrong as the ovary
ages?

Consequences of declining egg 'quantity' and egg 'quality'

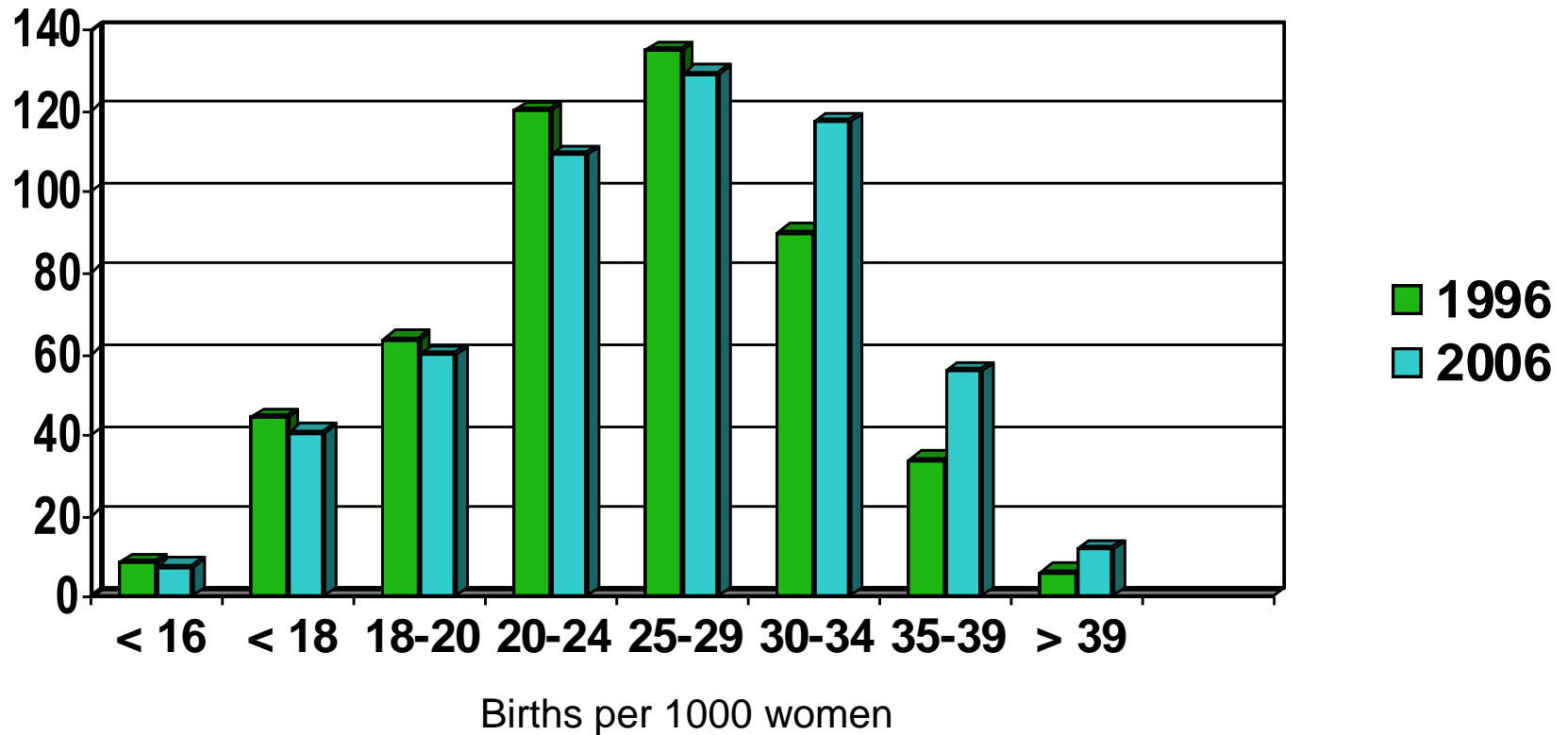


USA: Changing approaches to childbirth

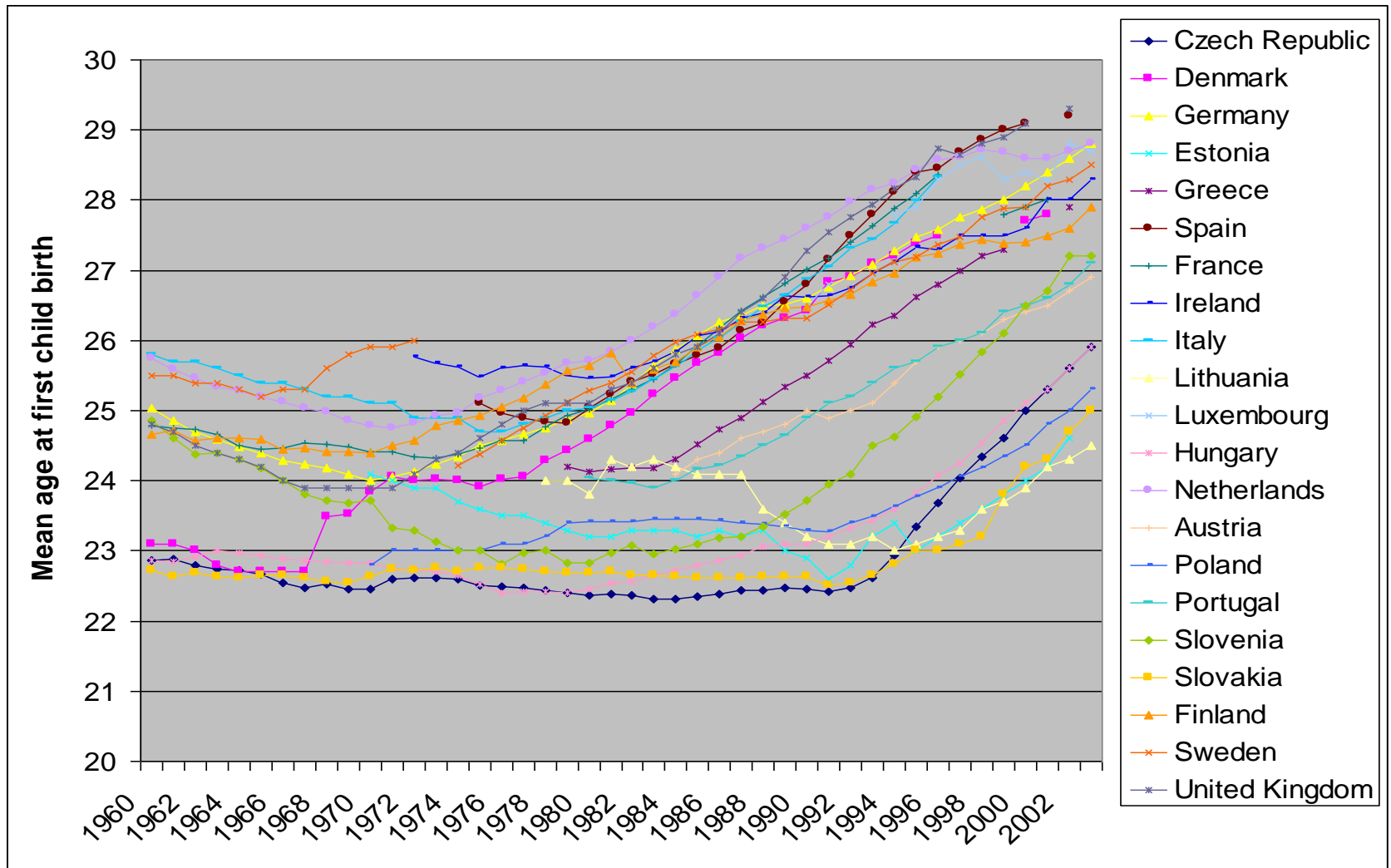
Age Birth Rate (%Change)



Delaying pregnancy, a modern phenomenon

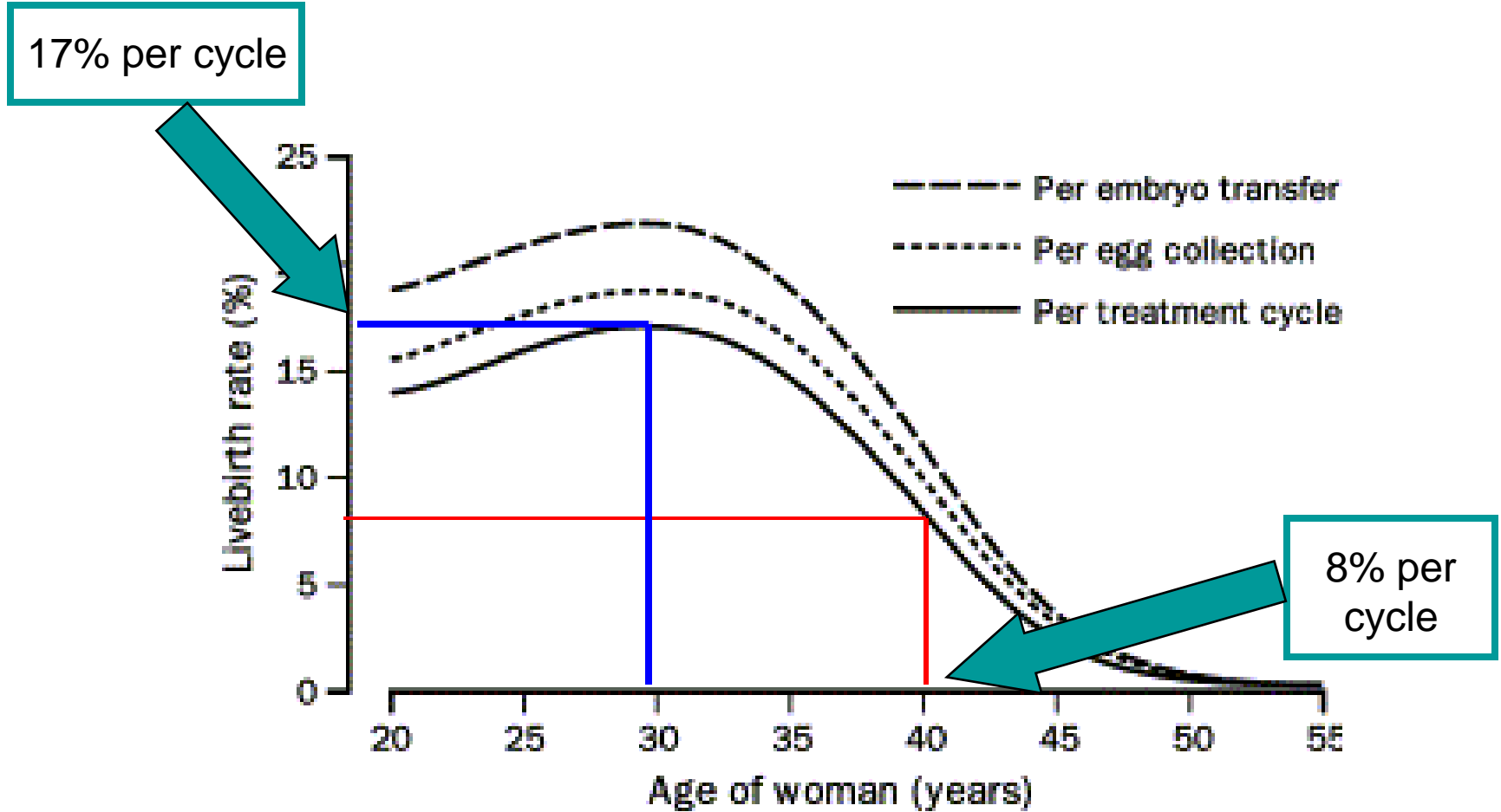


Mean age at first child birth



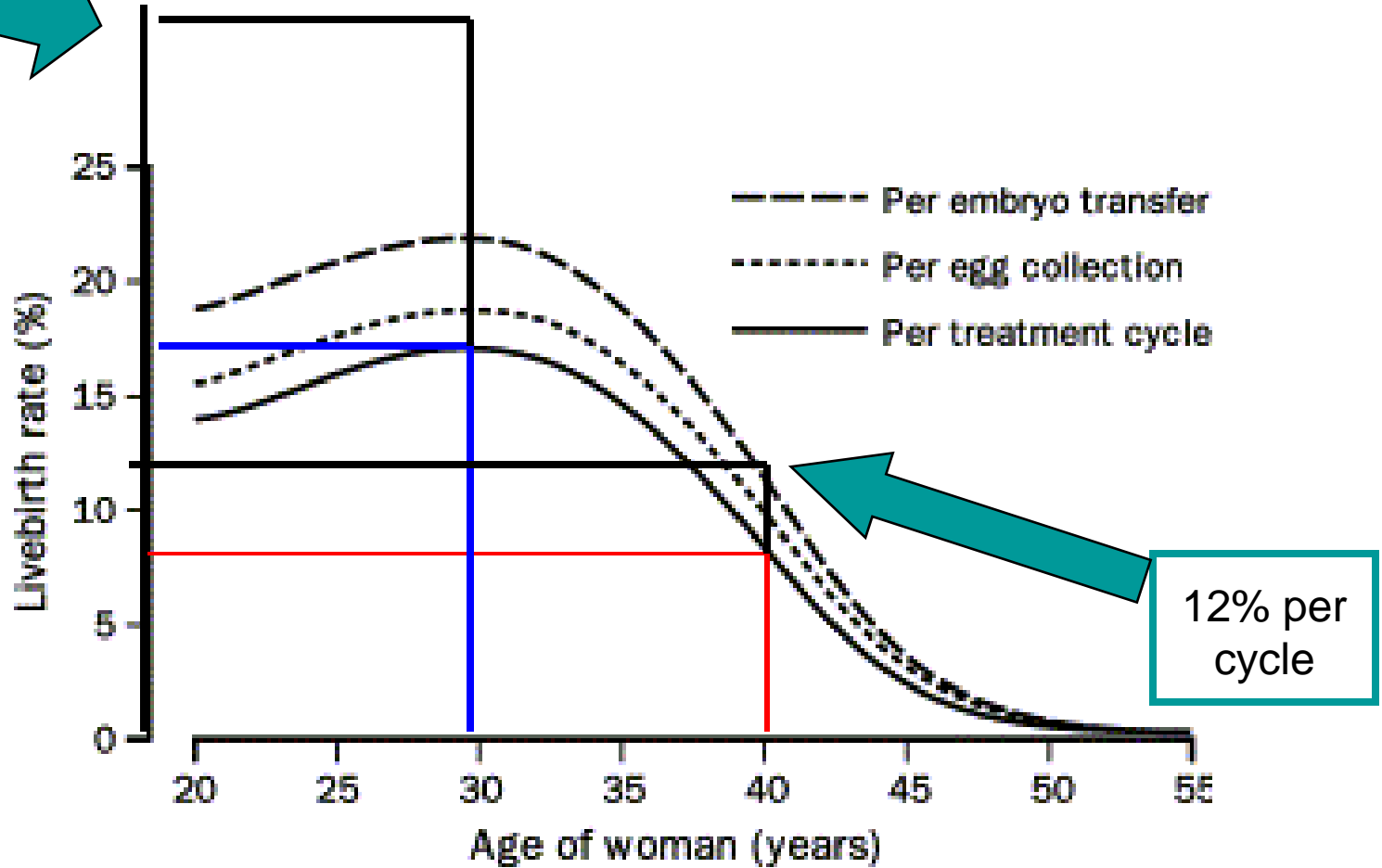
Can IVF solve the problem for the
older patient?

Not in 1994



31% per cycle

Not in 2010



NICE recommends.....

- Optimal female age range for IVF treatment is 23-39 years
- The chance of a live birth following IVF treatment is consistent for the first three cycles, therefore....
- Couples in which the woman is 23-39 years at the time of treatment and who have an identified cause for their fertility problems or who have unexplained infertility of at least three years duration should be offered up to three stimulated cycles of IVF treatment

UK cost effectiveness of IVF

- £2950 per cycle at age 30
- £3350 per cycle at 40
 - Higher requirement for gonadotropins
 - More monitoring
 - More cycle cancellations
- Cost per baby at age 30
 - £9033
- Cost per baby at age 40
 - £33500

But.... cost per baby for patients over 40

Cost per livebirth will increase with

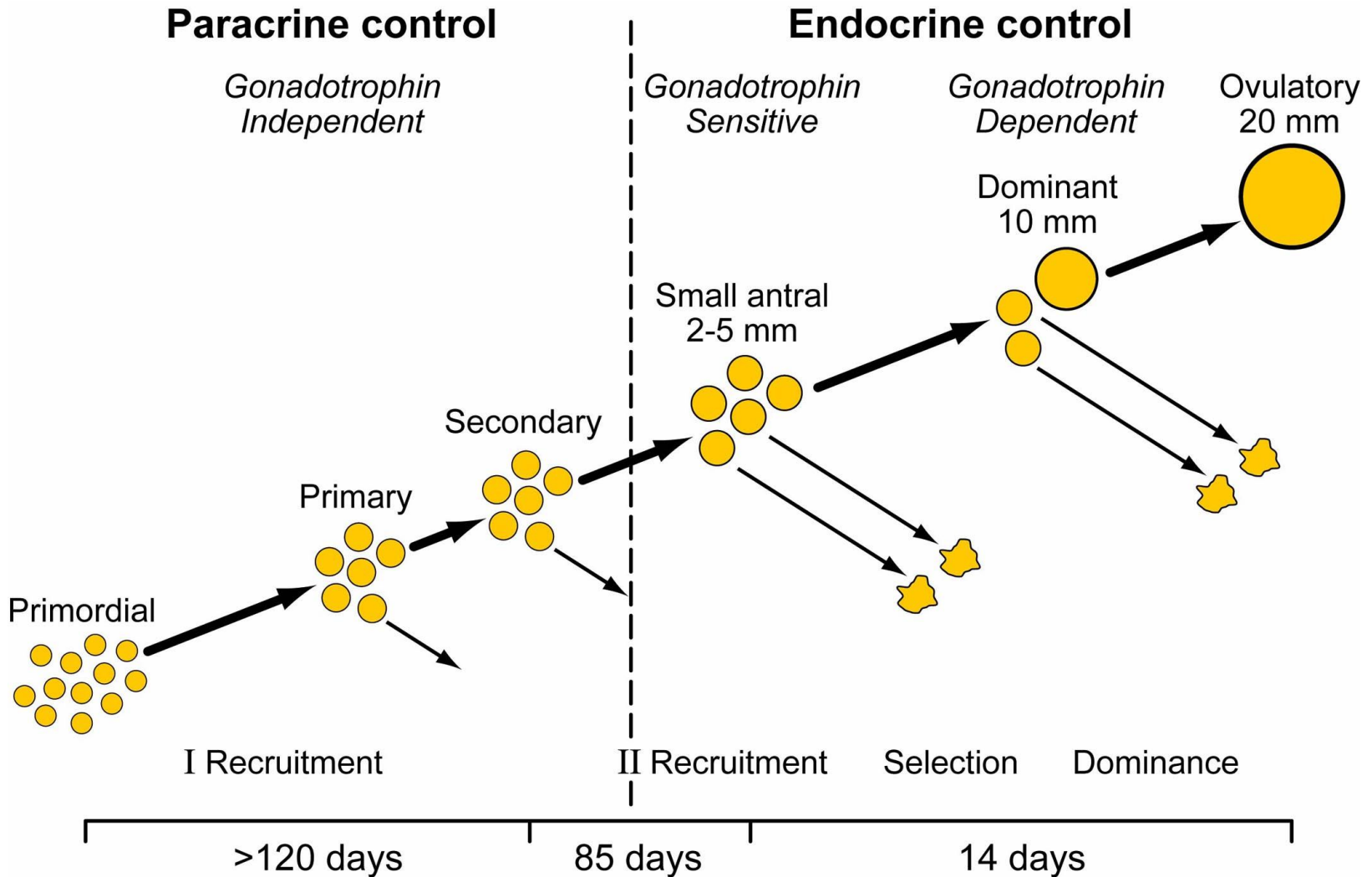
- higher doses of gonadotropins
- adjuvant therapies
- additional embryology costs
 - assisted hatching
 - preimplantation genetic screening

So how to treat the older patient?

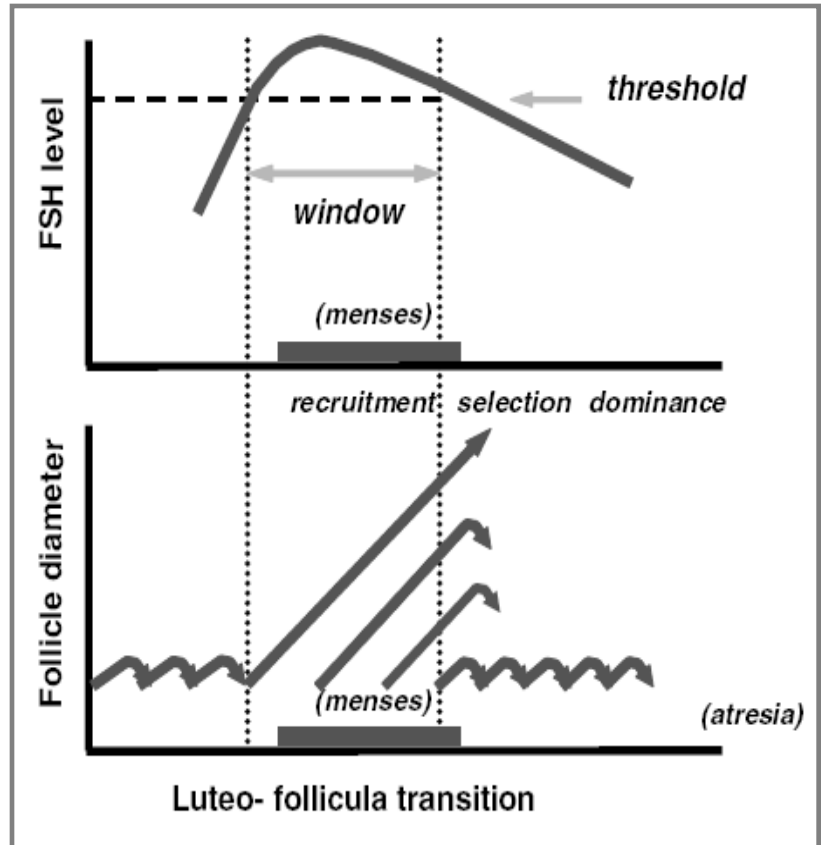
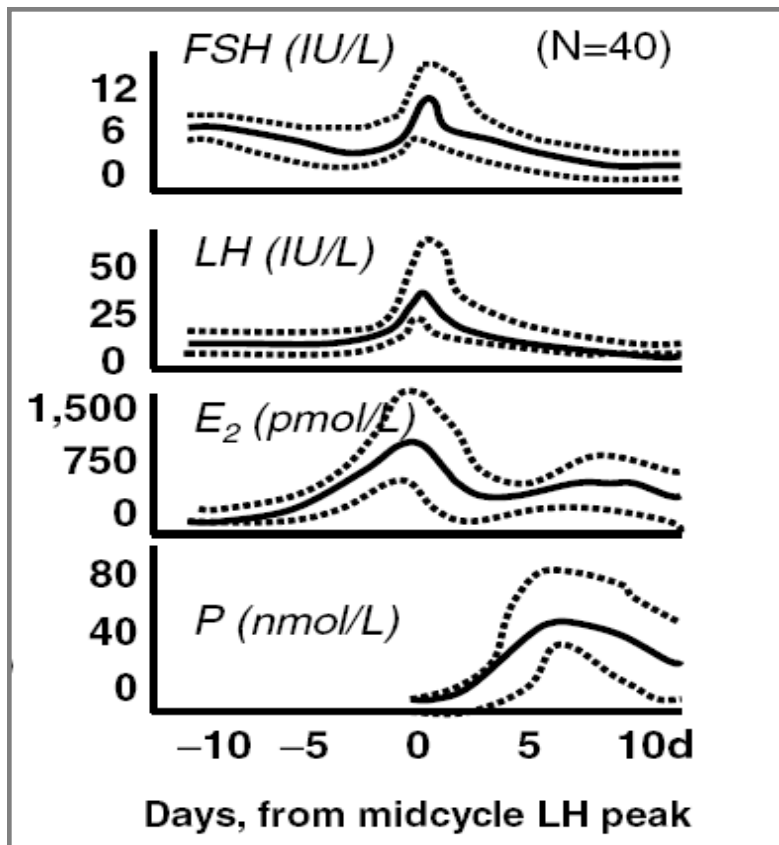
Improve the number of oocytes?

Heroic doses of gonadotropin

Follicle growth and maturation



Follicle recruitment and natural selection



So how to treat the older patient?

Improve uterine blood flow?

Sildenafil, aspirin

So how to treat the older patient?

Improve endometrial receptivity?

IVIg, heparin

So how to treat the older patient?

Improve embryo quality

Growth hormone

DHEA

Nuclear transfer

So how to treat the older patient?

Growth hormone

6 RCTs in poor responder patients. 169 patients

Greater proportion of patients reach embryo transfer

Increased live birth rates (rate difference: +17%, 95% CI: +5 to +30; fixed effects model) (NNT = 6; 95% CI: 3 - 20)

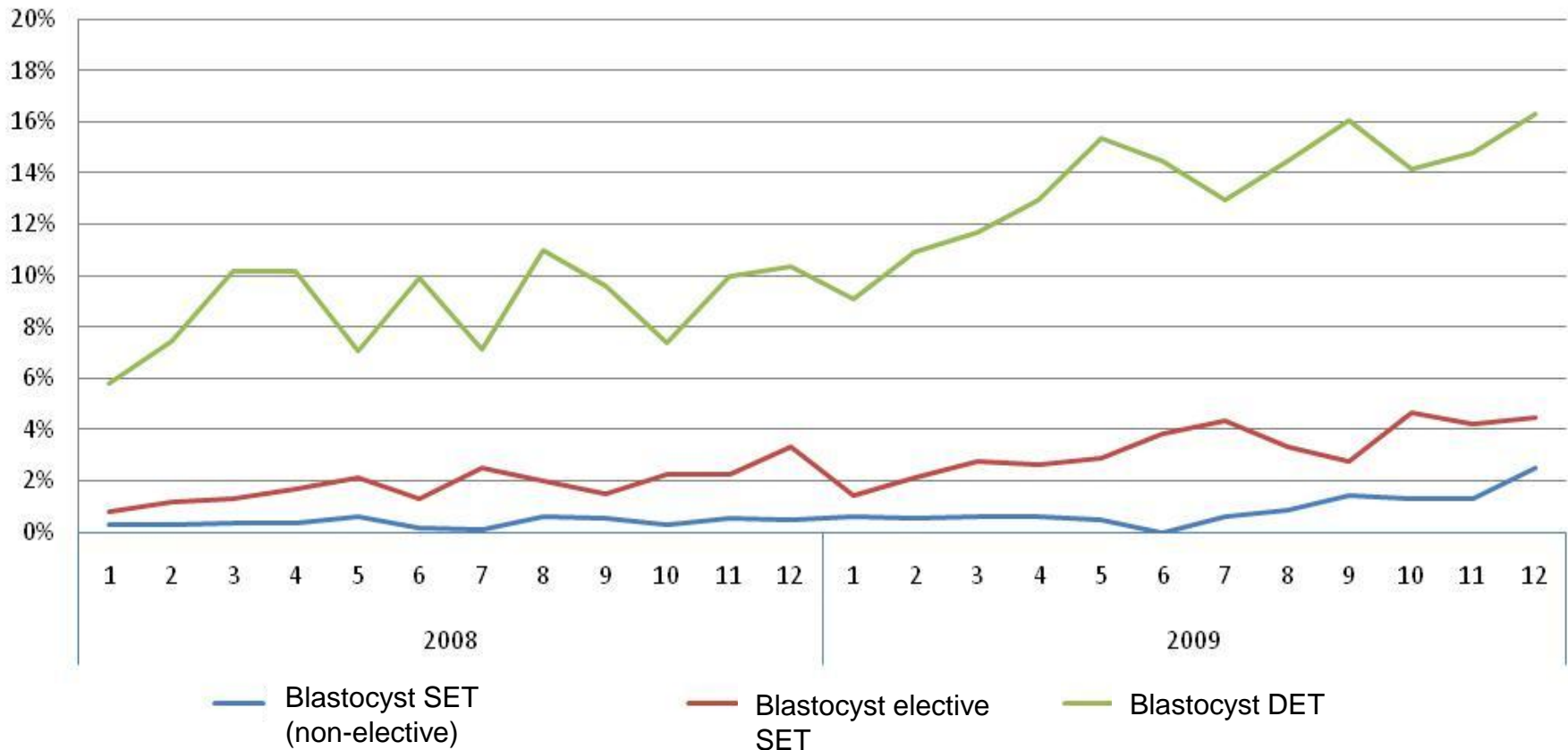
So how to treat the older patient?

Increase the 'hit' rate

Replace more embryos

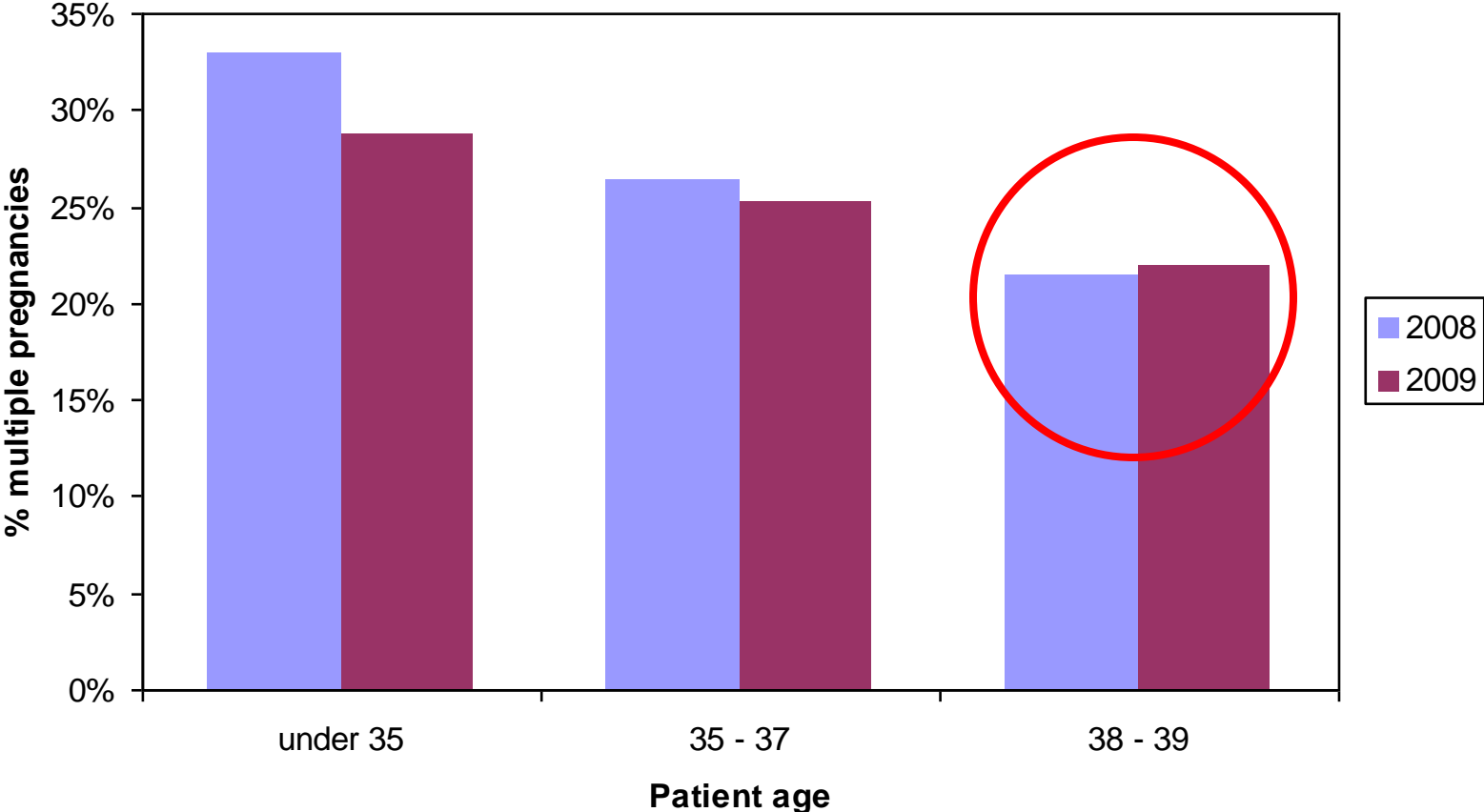
Double blastocyst transfer in women over 35

Blastocyst embryo transfers in women 35 - 39



Data extracted from HFEA Register 18/03/2010. 2009 data is unverified

Multiple pregnancies in 2009 compared to 2008



Data extracted from HFEA Register 18/03/2010. 2009 data is unverified

So how to treat the older patient?

Improve embryo quality?

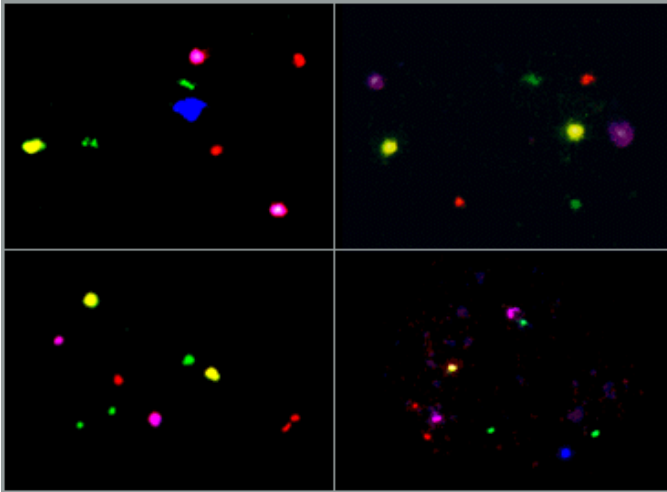
Preimplantation genetic screening

IVF as a means of accessing embryonic DNA

- Polar body biopsy
- Embryo biopsy
 - Cleavage
 - Blastocyst

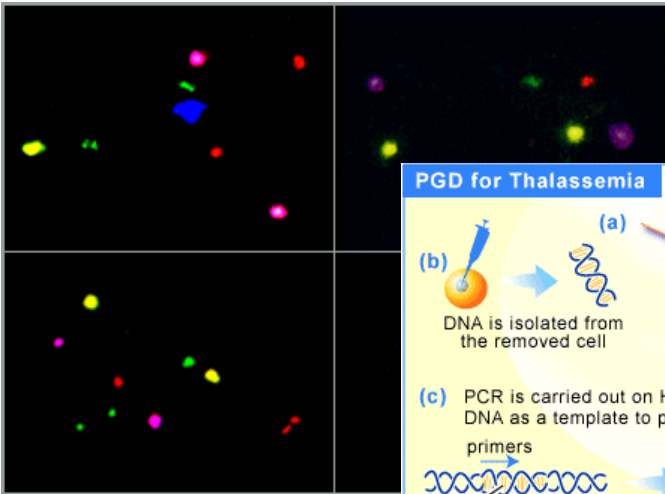
Genetic testing of a single blastomere

Chromosomal disorder/ sex selection FISH

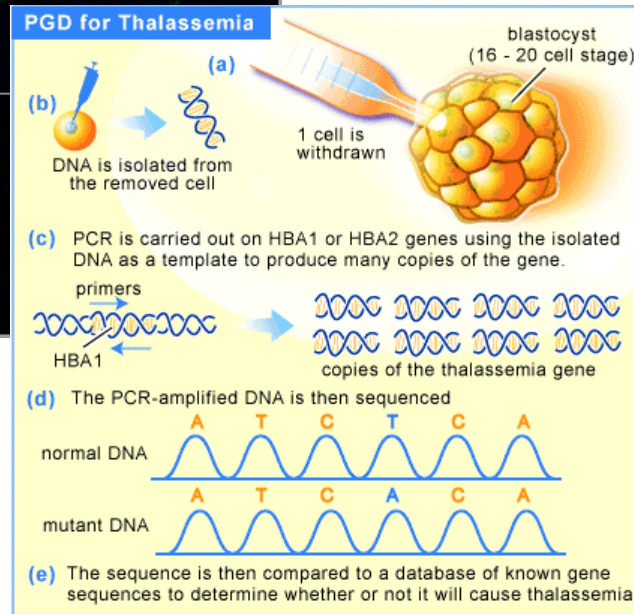


Genetic testing of a single blastomere

Chromosomal disorder/ sex selection FISH

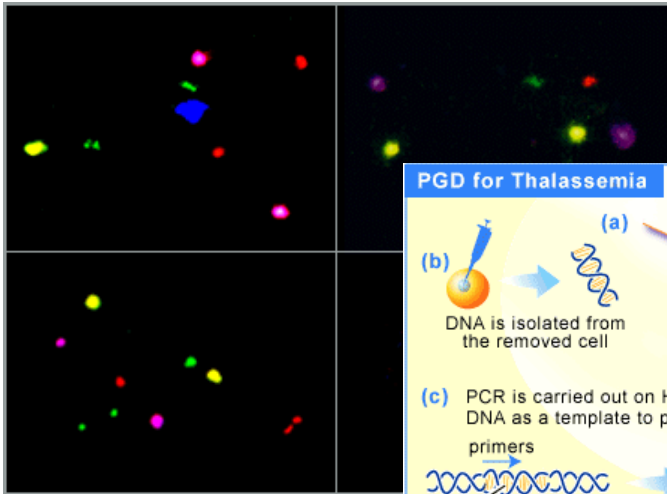


Single gene defect PCR

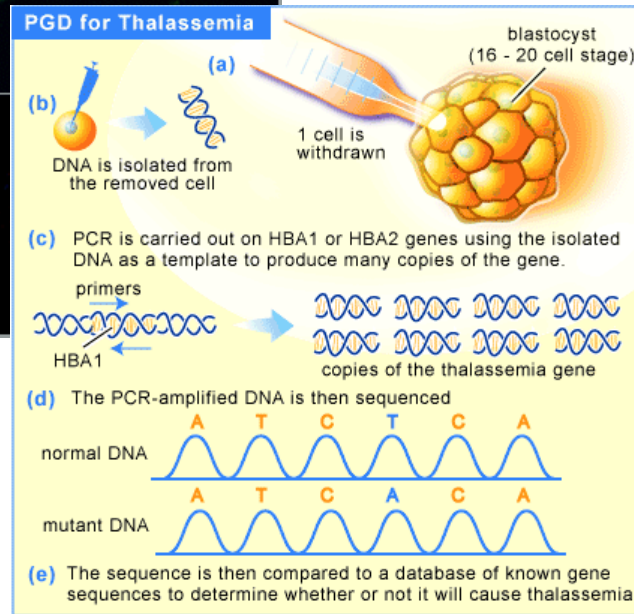


Genetic testing of a single blastomere

Chromosomal disorder/ sex selection FISH



Single gene defect PCR



The future - complete genomic hybridisation

So can we apply this
technology to aneuploidy
screening for older patients?

Pre-implantation genetic screening

The hypothesis

Embryos from older patients are frequently aneuploid

Aneuploidy cannot reliably be identified morphologically

PCR or CGH based technologies can be used to identify euploid embryos for transfer
This should improve pregnancy rates in older patients

Initial enthusiasm

Existing experience suggests that aneuploidy testing has had a significant impact on the reproductive outcome of poor prognosis IVF patients, which is based on the experience of over 20,000 cases of PGD for chromosomal disorders.

The clinical impact was demonstrated in the improved implantation and pregnancy rates, reduction of spontaneous abortions and improved take-home baby rate in patients of advanced reproductive age, those with repeated IVF failures and recurrent spontaneous abortions.

Kuliev & Verlinsky 2008

Reproductive Genetics Institute, Chicago

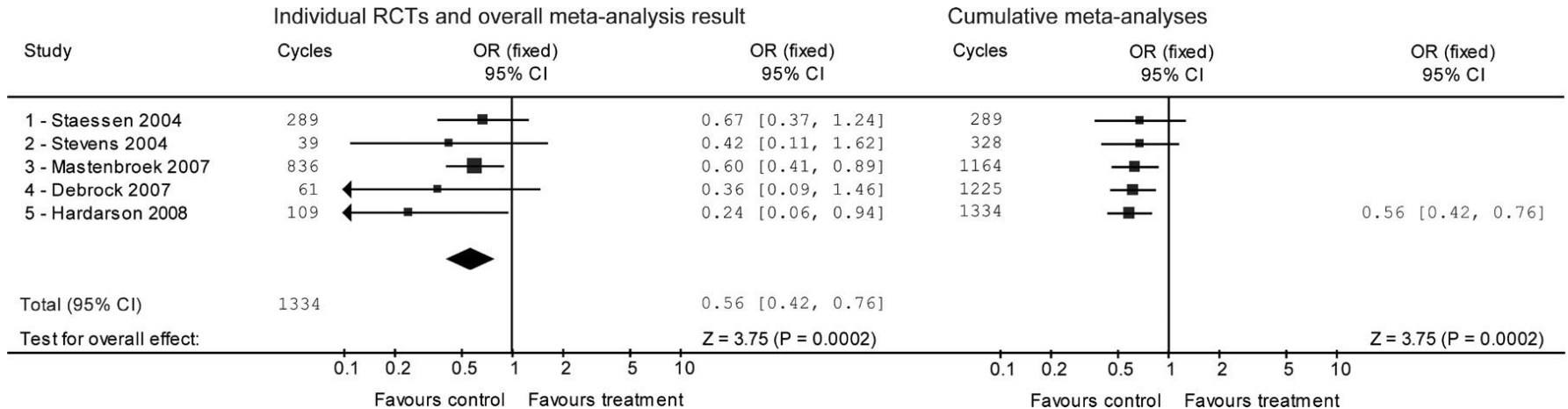
Despite the need for randomized controlled studies to quantify in more detail the clinical impact of the pre-selection of aneuploidy-free zygotes, the positive impact of PGD is particularly obvious from the comparison of reproductive outcome in the same patients with and without PGD, revealing the actual benefits of PGD

So what did the randomised trials show?

RCTs of pre-implantation genetic screening for older patients

reference	patients age	study design	conclusion
Staessen 2004	37	200 PGS vs 200 control Day 3 biopsy	No difference in embryo implantation or pregnancy rate
Mastenbroek 2007	35 - 42	206 PGS vs 202 control Day 3 biopsy	PGS lower pregnancy rate
Schoolcraft 2008	35	32 PGS vs 30 control Day 3 biopsy	No difference in implantation rate
Hardarson 2008	> 37	56 PGS vs 53 control Day 3 biopsy	PGS lower pregnancy rate
Jansen 2008	< 38	55 PGS vs 46 control Blastocyst biopsy	No difference in live birth per oocyte retrieval
Staessen 2008	< 36	107 PGS vs 107 control Day 3 biopsy	No difference in delivery rate

The effect of cleavage stage PGS in patients with advanced maternal age on ongoing pregnancy rate per cycle



Problems with PGS

- Potential for technical error in fixation & analysis
- FISH has 92 - 99% accuracy per probe
 - Multi probe techniques increase potential for error
- Mosaicism
- Reduced embryo survival
- Low numbers of embryos from older patients

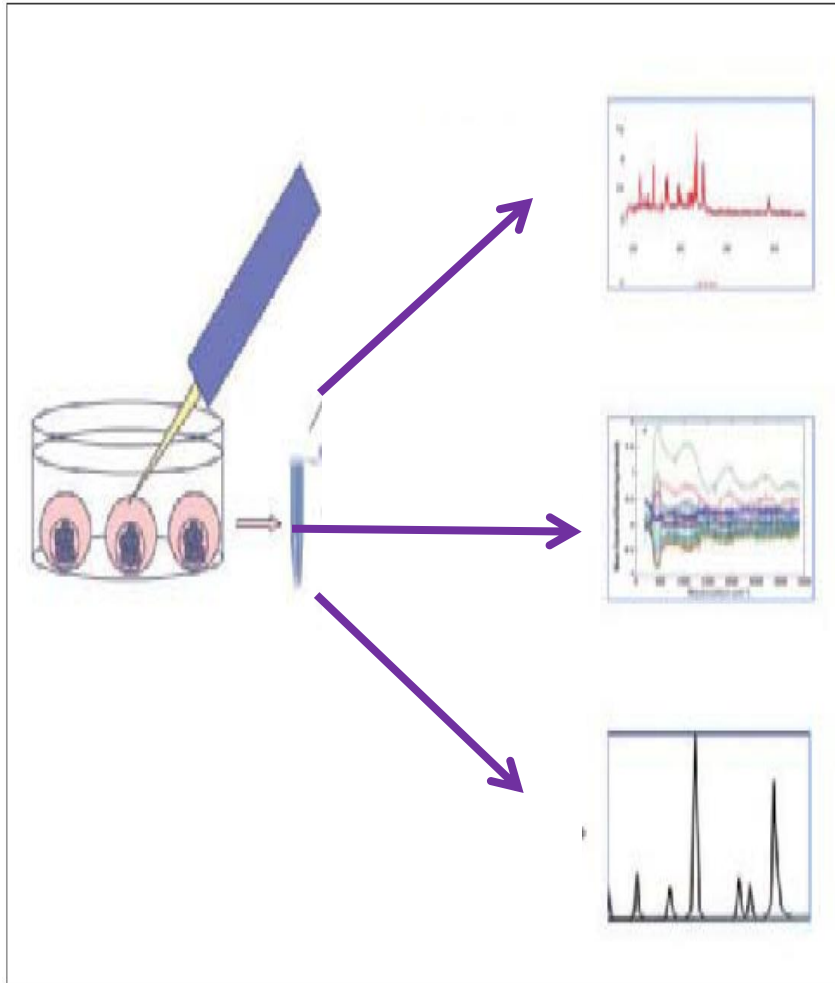
The numbers game

- Average yield of healthy embryos from one superovulated IVF cycle in a woman under 35 is ~ 10
- Fewer oocytes from older patients
- Techniques to select according to a specific genetic profile reduce the number available for transfer (NAT)
- Selection of multiple criteria sequentially reduces NAT even further
- One solution - artificial gametes from induced pluripotent stem cells (iPS) which could make thousands of embryos derived from one couple available for screening

Percentage of diploid and non-diploid blastomeres in human IVF embryos

	2 - 4 cell	5 - 8 cell	blastocyst
diploid	72%	59%	78%
aneuploid	28%	41%	22%

New technologies for embryo selection



Seldi-TOF Mass Spectrometry

- Correlation between day 5 secretome and blastocyst
- Ubiquitin key marker
- Panel of proteins being researched
- *Effects of media composition on secretome?*

Raman Spectrometry

- Allows analysis of multiple analytes
- Yields profile closer to range of secretions
- Predicted implantation in 80%

Scott R et al, FS 2008, 90:77

Nuclear magnetic resonance

- Metabolic profiling
- Sensitivity/specificity for pregnancy: 88%

Seli E et al FS 2008, 90: 2183

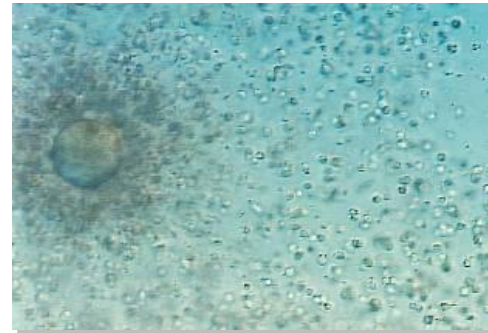
So how to treat the older patient?

'Store' fertility for later?

Egg or embryo freezing for young women?

Storage of oocytes (egg freezing)

- Requires an IVF cycle to superovulate and collect oocytes
- Takes 3 - 6 weeks
- Does not require a male partner
- “Slow” freeze or vitrification
- Fertilise later with ICSI



Slow freezing

- 2°C/min
- 0.3°C/min
- Ice crystal prevention by dehydration **during** cooling



Vitrification

- 15,000 – 30,000°C/min
- Ice crystal prevention by dehydration **before** cooling
- glass like state. No crystallisation (600 times faster)



Problems with oocyte freezing

- Medicalises healthy women
- Risks of procedure
- Uncertain chance of pregnancy later
 - False reassurance
 - Must freeze at a young age
- Risk to offspring
- Cost

So how to treat the older patient?

Use some-one else's eggs

Donor oocyte

Livebirth rate per embryo transfer following transfer of embryos from own or donated oocytes

	Live birth rate per embryo transfer	
Age	Own oocyte	Donor oocyte
< 29	23.7%	21.6%
30 - 34	21.9%	28.2%
35 - 39	17.6%	26.0%
40 - 44	8.1%	18.8%
> 44	3.5%	22.0%

Problems with oocyte donation for the older patient

- Probably will not have a suitable known donor
- Shortage of donors when payment prohibited and anonymity not available
- Ethics of reproductive tourism
 - Potential problems for donors
 - Short and long term
- Genetics
- Cost

How old is too old?

By EMMA MORTON
Health Editor
and GARY O'SHEA
Published: 16 May 2009

PROSPEROUS Elizabeth Adeney is to become a mum at 66 - so she has someone to "leave my money to".

The unmarried British businesswoman is almost eight months pregnant following IVF treatment. Her pregnancy stunned doctors, who warned that the birth poses massive health risks.

Elizabeth, of Newmarket, Suffolk, will take the title of Britain's oldest mum from doctor Patricia Rashbrook - who had a son at 63 in 2006

Rajo Devi, had been trying for 50 years to get pregnant with her 72-year-old [husband](#), who had failed to become a father in two prior marriages. It was undetermined whose egg and sperm were used in the treatment, the newspaper reported.

She became pregnant through in vitro fertilization at a clinic in the northern Indian state of Haryana

The worlds oldest new mother is dying at age 72 after giving birth just 18 months ago.

When she was 70, Rajo Devi Lohan and her husband Balla took out almost \$3,000 in loans for IVF treatments in Baddhu Patti, India, to conceive their only child, Naveen. Now 72, Rajo is bedridden and doesnt have the strength to lift 18-month-old Naveen. She admits she is dying, and too weak to recover from the pregnancy.

The future

- New approaches to PGS may overcome some technical problems
- But aneuploidy is not the only reason for poor IVF success rates with older patients
- Nuclear transfer to correct cytoplasmic defects?
- More use of donor oocytes?
- More oocyte cryopreservation?
- Or just have children at a younger age?