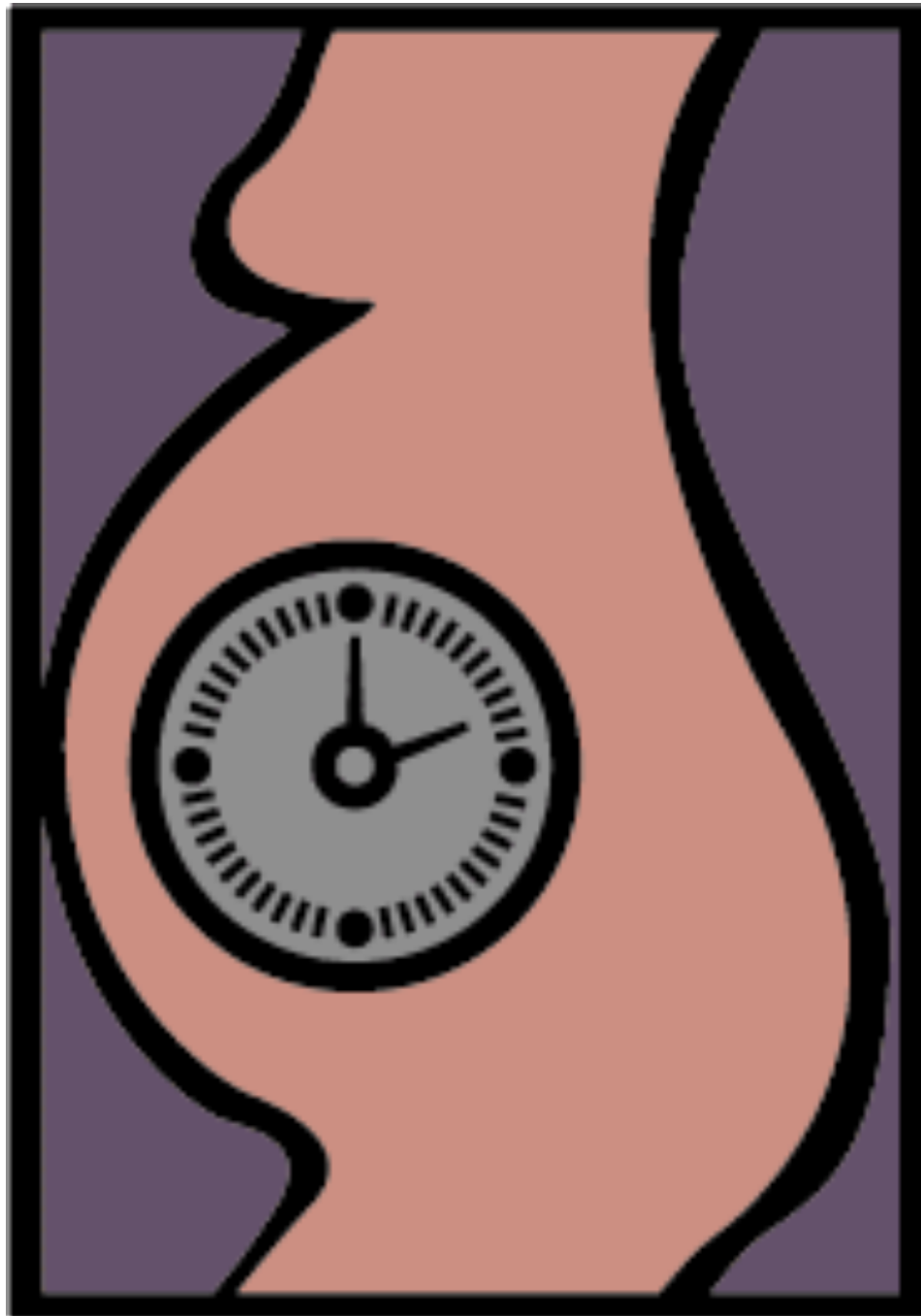


Environmental and Developmental Origins of Ovarian Reserve

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Why worry?

- Women delaying childbirth
- Diminished ovarian reserve major contributor to subfertility.
- A number of genetic loci associated with age at natural menopause. (Stolk et al 2012)
- Growing evidence of impact of developmental conditions and adult exposures.

'Fixed' link between reproductive milestones

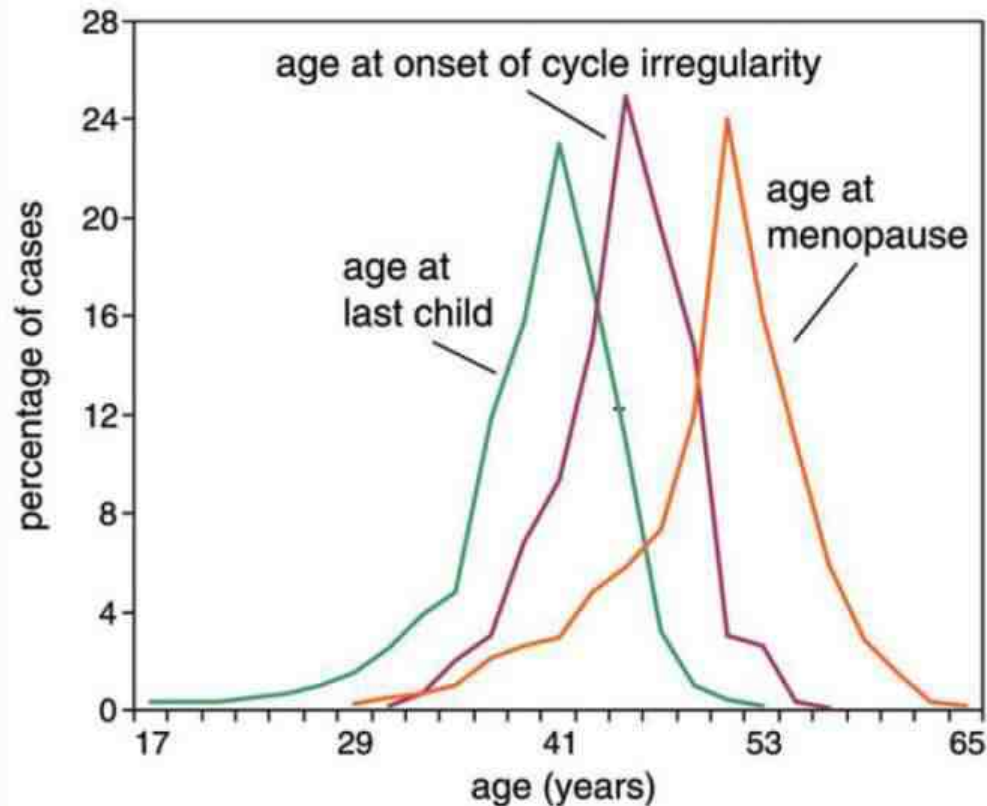


Figure 1 Variation with age with regard to age at last child, age at onset of cycle irregularity and age at menopause. Redrawn from Broekmans *et al.* (2009) with permission.

Programmed senescence?

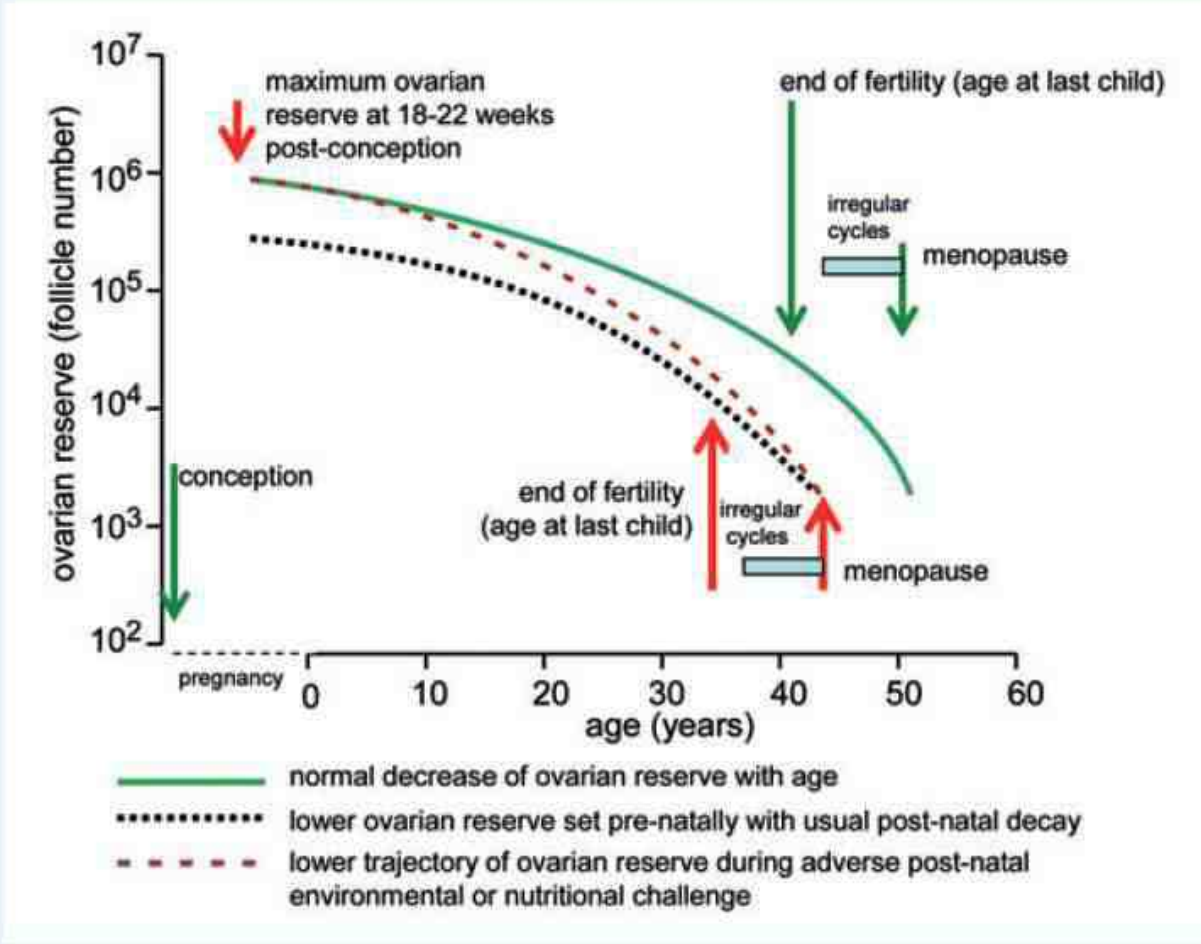


Figure 2 Decay of ovarian reserve with age.

Sou

DOHaD Concept of Programming



Malnutrition and other adverse environmental exposures during development alter gene expression and programme the body's structures and functions for life.

Can preconceptional nutrition, circulating hormones and maternal exposure during pregnancy influence ovarian reserve?

Intra-uterine nutrition and fetal reserve

- Maternal feed restriction during pregnancy in sheep:
 - smaller ovaries
 - fewer advanced follicles
 - no difference in germ cell numbers (Rae *et al.*, 2001)

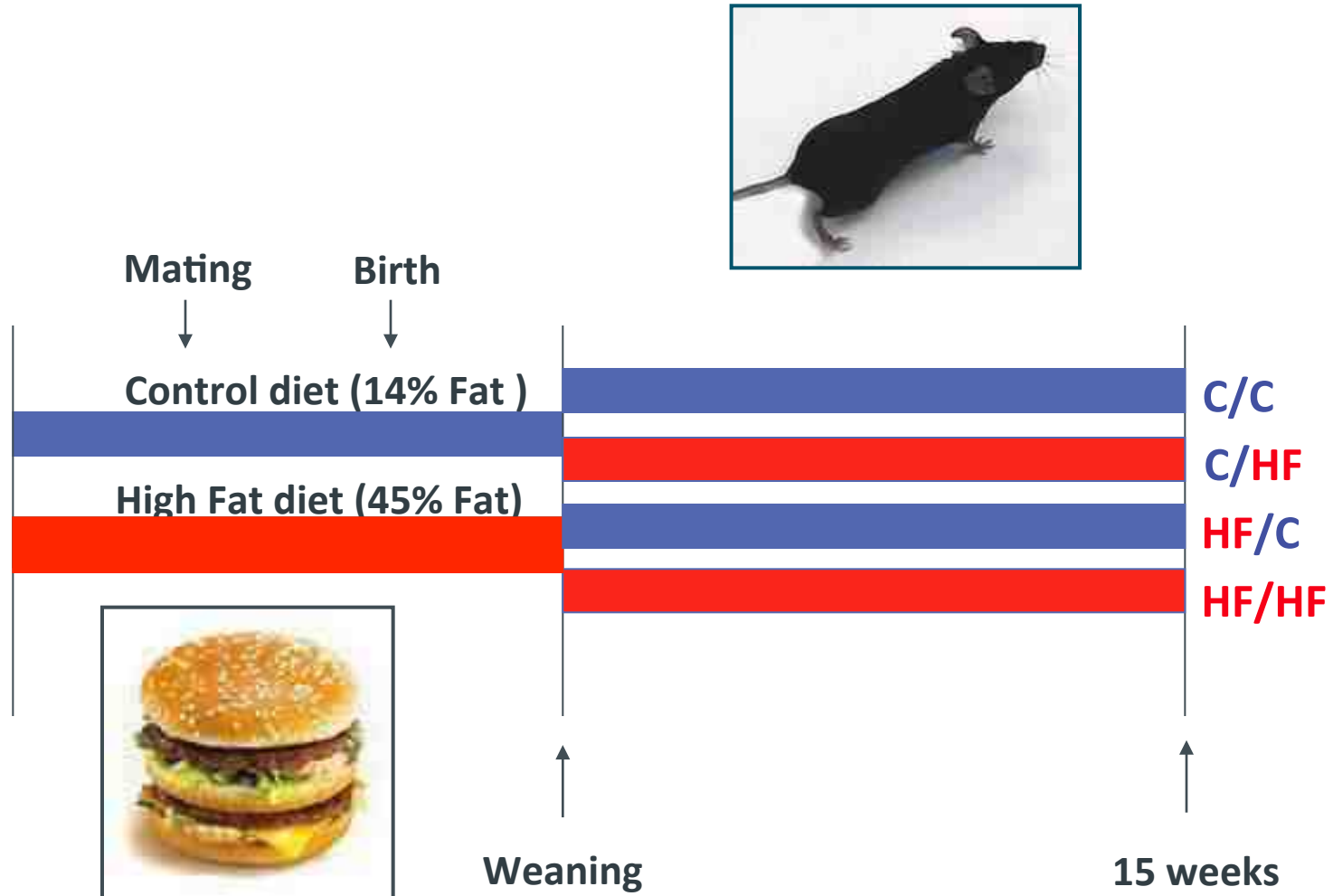
- 50% dietary reduction during gestation led to:
 - 23% fall in ovary weight without loss of germ cells

(Murdoch *et al.*, 2003)

→ Show poor (or delayed) follicular development without clear effects on overall oocyte numbers.



High fat periconceptional diet model



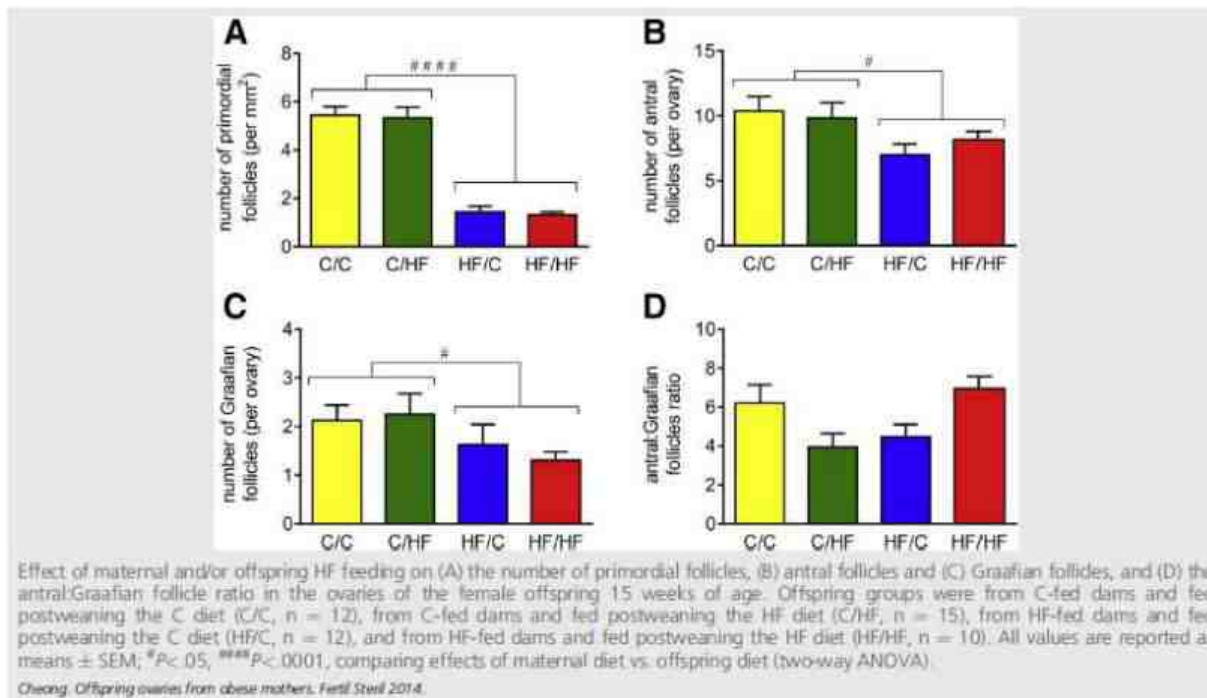
Diet-induced maternal obesity alters ovarian morphology and gene expression in the adult mouse offspring

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(Fertil Steril® 2014;102:899-907.)

FIGURE 2



Maternal obesity reduces primordial follicle pool...

Birthweight or early growth important?

- Extremes of birthweight (<2.5 kg or >4 kg) associated with an earlier age at menopause. (Tom *et al.*, 2010)
- Low weight gain in infancy is associated with an earlier menopause (Cresswell *et al.*, 1997).
- The importance of adequate lactation : maternal malnutrition during lactation shown to adversely affect follicular numbers in rats (Ferreira *et al.*, 2010).
- In humans, ovarian reserve varied positively with duration of breastfeeding and weight at 2 years, but not with birthweight (Hardy and Kuh, 2002).

Impact of prenatal endogenous androgens?

Table II Key studies and review linking prenatal exposure to androgens with later development of PCOS and associated effects on ovarian reserve.

Study	Key findings
Abbott <i>et al.</i> (2005)	Female rhesus monkeys treated prenatally with exogenous androgens develop features in adult life consistent with those observed in women with PCOS
Xita and Tsatsoulis (2006)	Evidence reviewed linking prenatal androgens with later development of PCOS
Steckler <i>et al.</i> (2005)	In sheep, <i>in utero</i> exposure to excess androgens causes a reduction in ovarian reserve through enhanced follicular recruitment
Dumesic <i>et al.</i> (2009)	In Rhesus monkeys, prenatal androgenization caused later development of decreased ovarian reserve, as measured through serum AMH and decreased oocyte yield in IVF cycles
Smith <i>et al.</i> (2009)	In sheep, evidence presented that much of the effect of prenatal androgens on ovarian reserve is through conversion to estrogens

PCOS, polycystic ovary syndrome; AMH, anti-Mullerian hormone.

But what about PCOS and Ovarian reserve?

- No evidence of earlier menopause
- Higher AMH (higher production per granulosa cell?)
- More follicles, but lower proportion at primordial stage
- Any reducing effect of prenatal androgens on reserve may be masked by ‘stockpiling’ of follicles by persistence and lack of ovulation.

Endocrine disruptors



- May interfere with hormone biosynthesis, signalling or metabolism
- Many act as androgen or estrogen receptor agonists/antagonists
- Can interfere with steroidogenesis and steroid metabolism.
- Residues detected in human serum, follicular fluid and seminal plasma.

Estrogen and Aryl hydrocarbon receptors are promiscuous 'open doors'. Ligands also affect oogenesis.

Endocrine disrupting chemicals (EDCs)

A range of mostly hydrophobic chemicals in the environment (derived from cigarette smoke, plasticisers, pesticides, cosmetics, dietary components, etc) are capable of interacting with the receptor systems below.

Polyaromatic hydrocarbons
(Smoking)

Bisphenols
Phytoestrogens (soya)
Pesticides

Aryl hydrocarbon receptor (AHR)

Estrogen receptor (ER)

Rapid response
involving membrane
associated ERs

Association with AHR nuclear
translocator (ARNT)

ER α

ER β

Genomic
pathway

Non-
genomic
pathway

Binding to dioxin
response elements
(DREs)

Binding to estrogen
response elements (EREs)

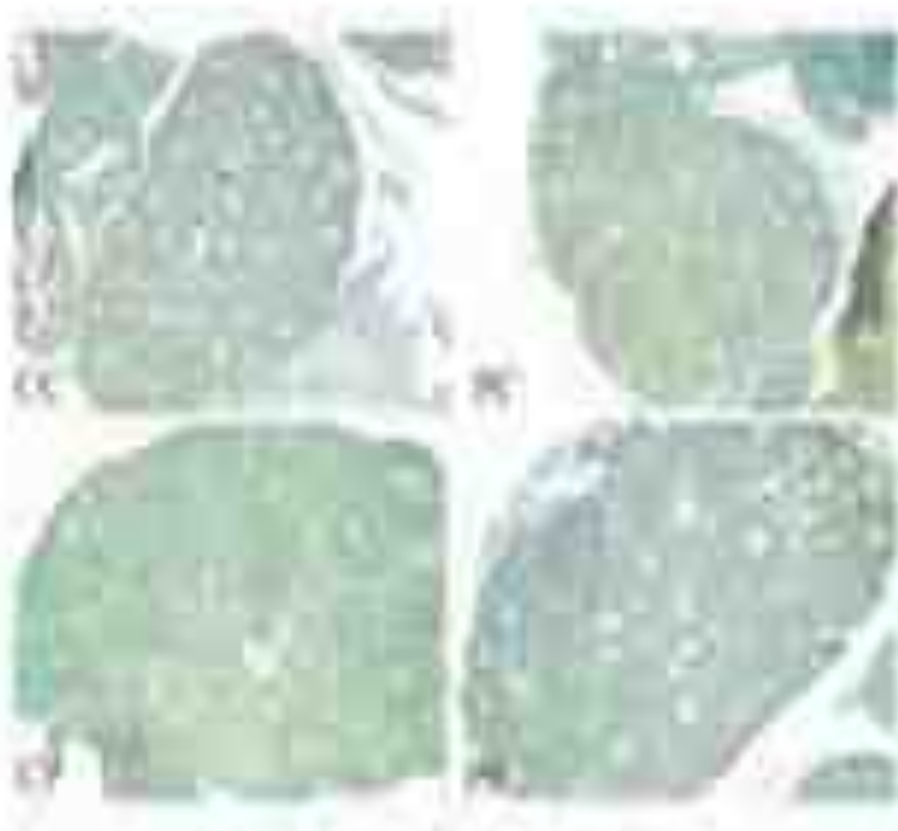
Kinase signalling
cascades





Maternal exposure to polycyclic aromatic hydrocarbons diminishes murine ovarian reserve via induction of *Harakiri*

Andrea Jurisicova,^{1,2} Asako Taniuchi,¹ Han Li,¹ Yuan Shang,¹ Monica Antenos,³ Jacqui Detmar,¹ Jing Xu,¹ Tiina Matikainen,⁴ Adalberto Benito Hernández,⁵ Gabriel Nunez,⁶ and Robert F. Casper^{1,2}



**Exposed ovaries:
Follicles depleted by two thirds**

**Xenotransplanted human ovary:
Same effect**

J. Clin. Invest. **117**:3971–3978 (2007)

Maternal Smoking and Male Fertility

- 24 human first-trimester testes, aged 37-68 days post-conception, obtained from women undergoing legal termination of pregnancy . Historical controls.
- Testes exposed to maternal smoking:
- reduction in the number of germ cells: 55% [74-21%] $P = 0.004$
- Reduction in number of somatic cells by 37% [59-3%] $P = 0.023$
- The effect of maternal smoking was dose-dependent
- The number of germ cells in embryonic gonads, irrespective of gender, was also significantly reduced by 41% (95% CI 58-19%, $P = 0.001$) in exposed versus non-exposed embryonic gonads.

Little controversy...

Table III Key studies and reviews linking cigarette smoking with the loss of ovarian reserve.

Study	Key findings
Harlow and Signorello (2000)	Evidence summarized indicating that smoking hastens the onset of the menopause by as much 1–2 years
Sharara <i>et al.</i> (1994)	Smoking accelerates the development of diminished ovarian reserve as evidenced by poorer FSH responses during the 'Clomiphene Citrate Challenge Test'
Cooper <i>et al.</i> (1995)	Active and passive smoking are associated with elevated FSH concentrations in women 38–49 years old
Zenzes <i>et al.</i> (1997)	Smoking reduces number of mature oocytes retrieved following ovulation induction in IVF cycles
El-Nemr <i>et al.</i> (1998)	Raised basal FSH and lower numbers of oocytes retrieved in IVF cycles in smokers compared with non-smokers
Fuentes <i>et al.</i> (2010)	Recent smoking is significantly associated with a decreased number of retrieved ova in IVF cycles
Waylen <i>et al.</i> (2009)	General review of the effect of smoking on the outcomes of ART. Poorer ovarian reserve is likely to be an important factor in lower pregnancy rates in smokers following ART

ART, assisted reproduction technologies.

Bisphenol

- One of highest volume chemicals produced
- Used in synthesis of plastics, inner coats of cans
- Can leach out and contaminate food
- Fits into ER binding pocket
- May act via membrane bound ERs, triggering non-genomic mechanism

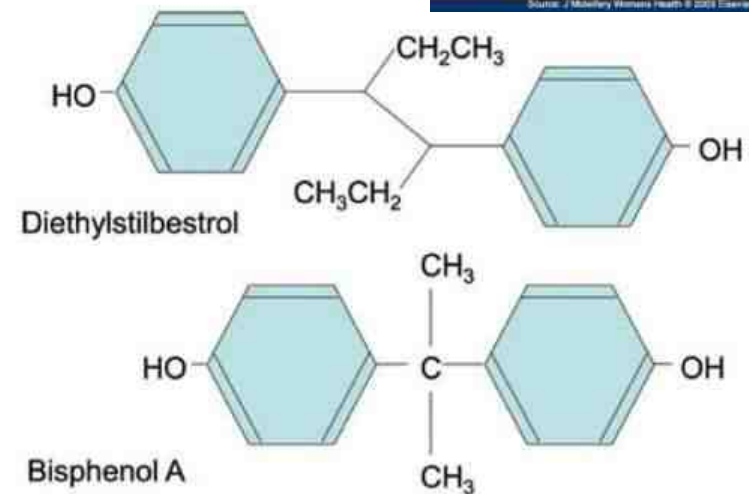


Figure 5 Structural similarity of diethylstilbestrol (DES) and bisphenol A (BPA). DES is a pharmaceutical which binds with high affinity to the ER, whereas BPA is used in the synthesis of plastics and binds to the ER with low affinity.

BPA and oocyte quality: meiotic abnormalities

Table IV Key studies linking the exposure to BPA with development of meiotic abnormalities in oocytes.

Study	Key finding
Hunt <i>et al.</i> (2003)	Exposure of female mice to BPA released from damaged plastic caging material and water bottles causes increased aneuploidy in their oocytes
Can <i>et al.</i> (2005)	BPA induces delay in the meiotic cell cycle altering centrosome and spindle microtubular organization
Lenie <i>et al.</i> (2008)	Exposure of mouse follicular cultures to BPA causes meiotic abnormalities including changes in spindle formation
Mlynarcikova <i>et al.</i> (2005)	BPA causes alterations in steroid production by ovarian granulosa cells
Susiarjo <i>et al.</i> (2007)	Exposure of pregnant mice to environmentally relevant doses of BPA caused meiotic abnormalities in the oocytes within the fetal ovaries. The ovaries of the resulting female mice contained oocytes with increased rates of aneuploidy
Mok-lin <i>et al.</i> (2010)	BPA is detectable in the majority of women undergoing IVF and is inversely associated with the number of oocytes retrieved

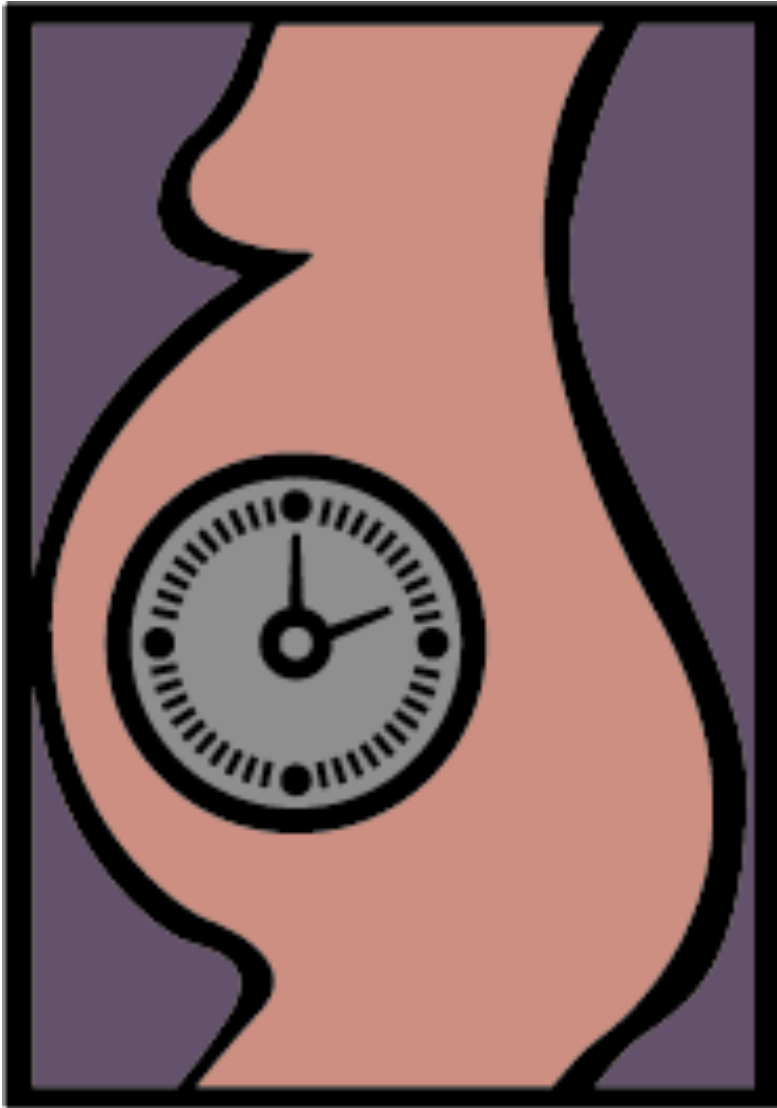
BPA, bisphenol-A.



Socioeconomic factors: Education

- Review of 29 studies: weak association between educational attainment and later ANM (Canavez et al 2011)
- Link between cognitive ability in childhood and later ANM (Richards et al 1999)

Confounding variables?



Multiple determinants

**Developmental and
environmental effects
may be small**

But they can be influenced.

Conclusions: Early Life Influences

- Role of nutrition unclear, but population studies indicate role.
- Convincing case for Endocrine Disruptors
- Good evidence for prenatal androgen exposure contributing to aetiology of PCOS and diminished ovarian reserve: perhaps by estrogen actions after aromatisation of androgens
- BPA accumulates in amniotic fluid: may affect androgen related events important in establishing ovarian reserve

Conclusions: Adult Influences

- Poor nutrition associated with fall in ANM.
- No evidence that excess fat leads to loss of ovarian reserve.
- Good evidence that smoking reduces ANM.
- Mechanisms include pro-apoptotic effects of PAHs working via AHR.
- Disrupting effect of BPA on meiotic maturation of oocytes, causing aneuploidy and cell cycle arrest.

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human
reproduction
update

Environmental and developmental origins of ovarian reserve

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