Involvement of local factors in aberrant follicle development

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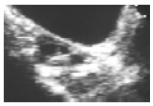
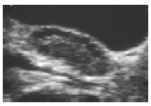
Involvement of local factors in aberrant follicle development

- Follicle development in the human ovary
- Disordered preantral follicle development in PCOS
- Local factors implicated in aberrant follicle development in PCOS

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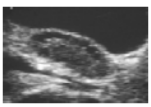
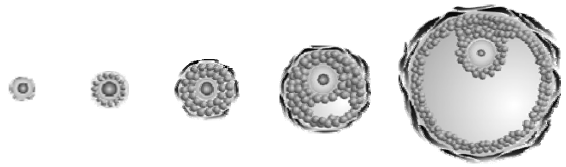


Human follicle development

Preantral	Antral
Gonadotropin "sensitive" (several months)	Gonadotropin dependent (six weeks)

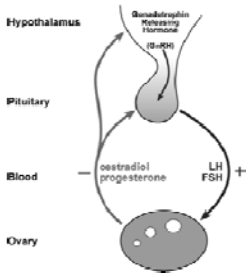


40 µm	50 µm	>70 µm	0.2 mm	20 mm
primordial	primary	secondary	early antral	Graafian (pre-ovulatory)

Anovulation is a common cause of infertility

- Disorders of ovulation account for about 25% of causes of infertility
- Most are due to abnormal endocrine environment
- Most are treatable

Causes of anovulation



- Primary ovarian failure (8%)
- Deficiency or disordered regulation of gonadotrophins (32%)
  - Functional: weight loss; exercise; idiopathic
  - Organic: Kallmann's syndrome and its variants; prolactin excess
- Polycystic ovary syndrome (55%)
- Miscellaneous (5%)

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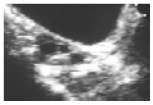
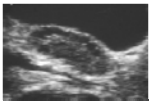
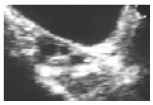
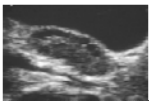



Polycystic ovary syndrome

- Characterised by anovulation with clinical (hirsutism/acne) and/or biochemical evidence of androgen excess
- Typically presents during adolescence
- Affects >5% women of reproductive age
- Commonest cause of anovulatory infertility (>75% cases)
- Typical endocrine features are raised testosterone and LH
- Also associated with metabolic abnormalities and increased risk of type 2 diabetes

Polycystic ovary syndrome

- Aetiology uncertain
- Genetic factors important

Developmental origin of PCOS: an hypothesis

- Polycystic ovary syndrome is a genetically-determined, primary ovarian disorder resulting in excess androgen production
- The capacity to hypersecrete androgens begins in fetal life
- Typical clinical and biochemical features of PCOS are "downstream" effects of exposure to androgen excess at or before puberty
- Phenotype may be influenced by other genes and by environment

Abbott, Dumesic & Franks, *J Endocrinol* 2002, 174 1-5

A primate model for PCOS: evidence for a key role for androgens

Rhesus monkeys, exposed to high concentrations of androgens during fetal life, as adults show:

- Hypersecretion of LH
- Ovarian hyperandrogenism
- Insulin resistance
- Anovulation in relation to increased body weight

Abbott *et al, Trends Endocrinol Metab* 1998 **9** 62-7
Eisner *et al, J Clin Endocrinol Metab* 2000 **85** 1206-10
Eisner *et al, Fertil Steril* 2002 **77** 167-72

(Also sheep and rodent models)

Developmental origin of PCOS

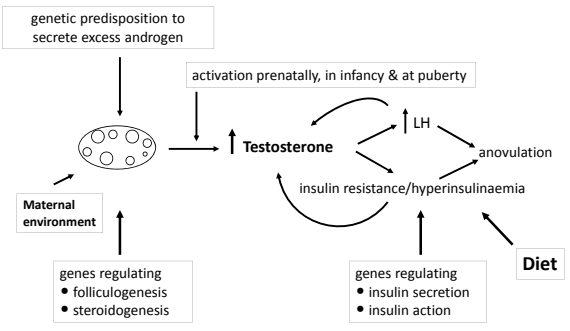
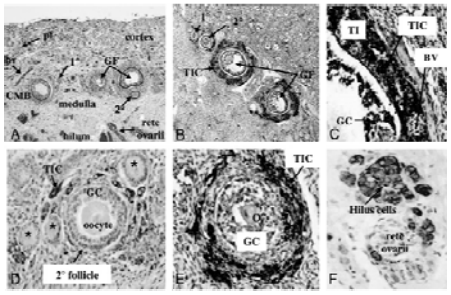


FIG. 2. Analysis of control human fetal ovaries during the third trimester: morphology and immunohistochemical localization of P450c17



Cole, B. et al. J Clin Endocrinol Metab 2006;91:3654-3661

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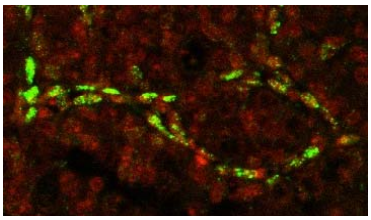
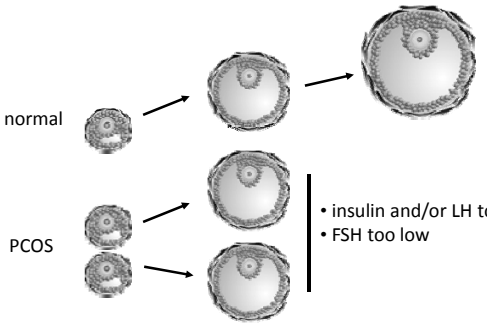
Androgen receptor expression in 19w human fetal ovary



AR+ve somatic cells surround clusters of germ cells

Richard Anderson; from Fowler *et al*, Development of Steroid Signaling Pathways during Primordial Follicle Formation in the Human Fetal Ovary *J Clin Endocrinol Metab*, 2011 96 1754-62

Arrested antral follicle development in PCOS



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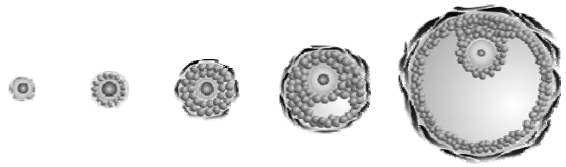

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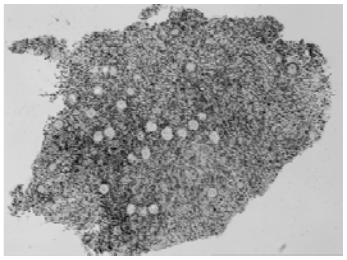
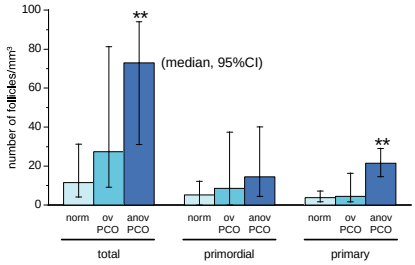
What about preantral follicle development in PCOS?

Analysis of biopsies of ovarian cortex

- Biopsies of ovarian cortex, fixed, serially sectioned & stained (H&E)
- Follicles:
  - counted
  - assessed for stage of development &
  - atresia/health

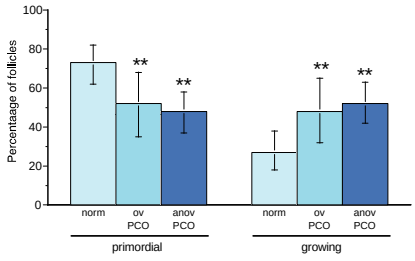


Preantral follicle density increased in PCO



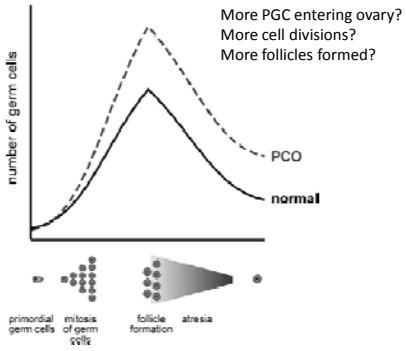
Webber *et al* Lancet 362, 1017 (2003)

Increased proportion of growing follicles in polycystic ovaries

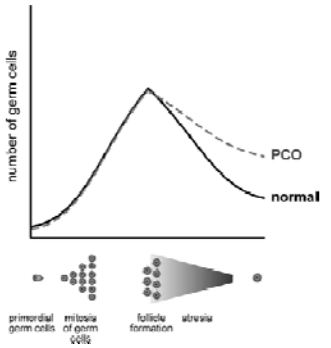


Webber *et al* Lancet 362, 1017 (2003)

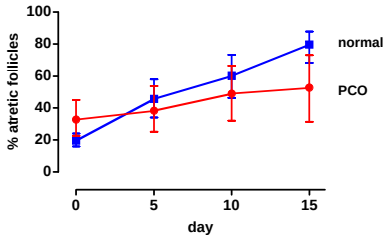
Increased density of preantral follicles: higher initial population of primordial follicles?



Increased density of preantral follicles: lower rate of atresia in PCO?

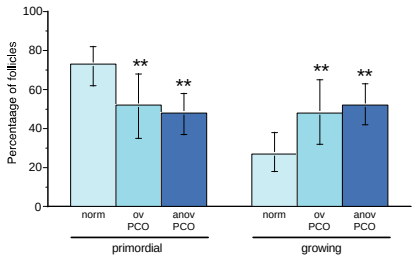


Increased survival of preantral follicles from PCO in culture



Webber *et al, J Clin Endocrinol Metab* 2007 **92** 1975-8

Increased proportion of growing follicles in polycystic ovaries

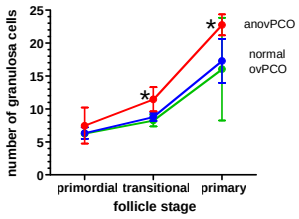


Webber *et al* Lancet 362, 1017 (2003)

Reduced atresia may contribute to the higher density of preantral follicles in PCOS

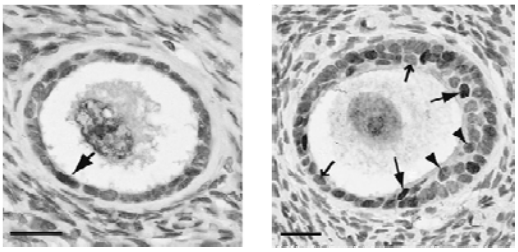
But what factors are involved in increased activation?

More granulosa cells in PCO



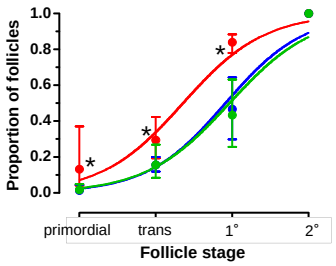
Quantitative histology using sections of archived human ovaries
Stubbs SA, et al. 2007; *J Clin Endocrinol Metab* 92: 4418

MCM-2 (cell proliferation marker) in primary follicles

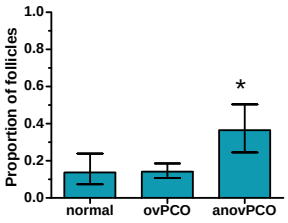


Stubbs SA, et al. 2007; *J Clin Endocrinol Metab* 92: 4418

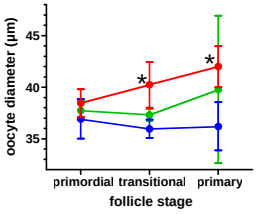
Increased proportion of MCM2-positive follicles



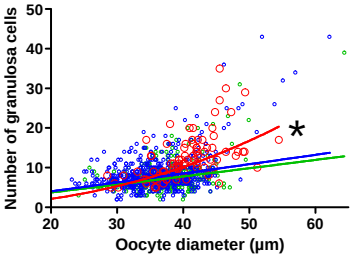
Increased proportion of MCM2-positive preantral follicles

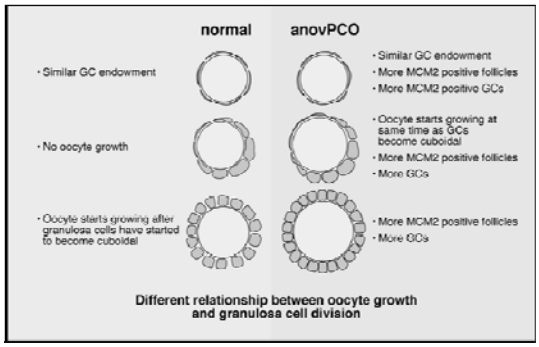


Larger oocytes in PCO



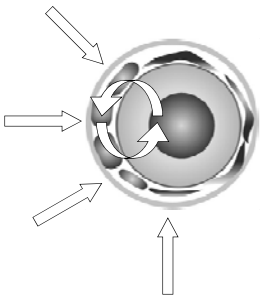
Altered relationship between oocyte growth and GC proliferation





Implications:

- Intrinsic defect in early follicle development in PCO, which may involve
  - altered inter-follicular signalling
  - altered intra-follicular signalling



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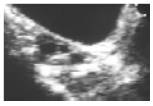
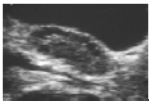
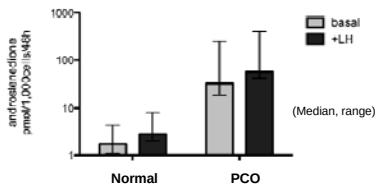



Increased activation of primordial follicles in PCO

- less inhibitor?
  - Reduced AMH expression in early preantral follicles
    (Stubbs et al, 2005)
- increased stimulator?
  - IGFs
  - androgens

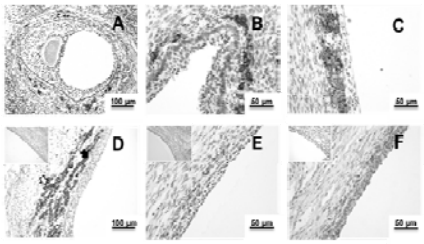
Source of excess androgen in PCOS



PCO theca cells produce 20-fold more androstenedione in culture than normal theca; phenotype is maintained after several passages

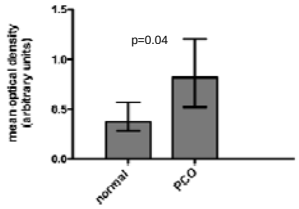
Gilling-Smith *et al*, 1994, Nelson *et al*, 2001

P450c17 protein expression in human antral follicles



Fabio Comim

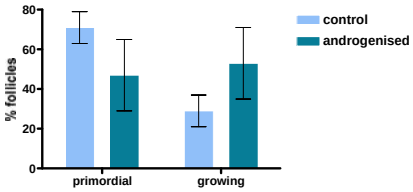
Expression of P450c17 in theca of normal and polycystic human ovaries



Androgens and preantral follicle development

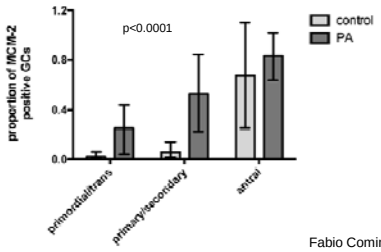
Lessons from the prenatally andogenized sheep

Initiation of follicle development in PA sheep (8 months)



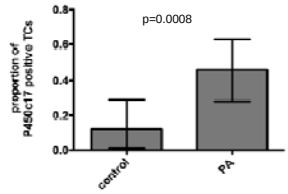
Forsdike RA, et al. 2007; *J Endocrinol* 192: 421

Enhanced granulosa cell proliferation (MCM2 +ve) in prenatally androgenized sheep ovary

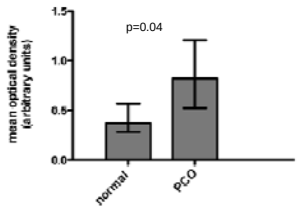


Fabio Comim

P450c17 expression is increased in theca of prenatally androgenized sheep ovary



Expression of P450c17 in theca of normal and polycystic human ovaries



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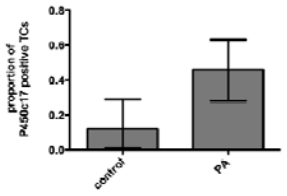
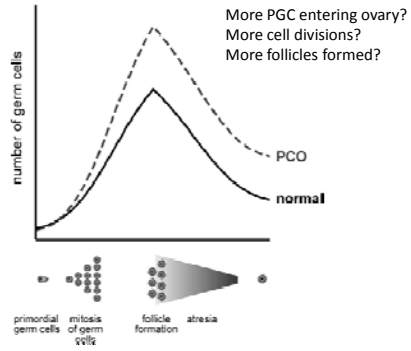
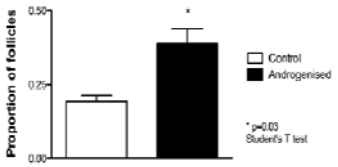


Implication: androgen "programming" leads to increased androgen production by theca (similar data in Rhesus)

Increased density of preantral follicles: higher initial population of primordial follicles?



Proportion of germ cells in follicles in fetal sheep ovary (d90)



Androgens and aberrant follicle development in PCOS

- Prenatal exposure to excess androgen in sheep affects follicle formation and early follicle development
- Is this relevant to PCOS?
  - AR in fetal and adult follicles
  - abberant follicle development shows many features similar to effects of PA in sheep
- Where does excess androgen come from in human PCOS, what causes it and when is it manifest?

Summary

- Preantral follicle development depends largely on local ovarian factors
- Preantral follicle development is abnormal in PCOS
- The local factors involved remain unclear but exposure to excess androgen in early life may be a key event

What comes first - abnormal early follicle development or increased ovarian androgen production?

With special thanks to…

- Fabio Comim
- Kate Hardy
- Lisa Webber
- Sharron Stubbs
- Rachel Forsdike
- Jane Robinson
- Jaroslav Stark

Imperial College London

CAPES (Brazil)

MRC Medical Research Council