In vitro follicle growth to investigate follicular paracrine interactions –

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Please Note: These slides may not represent the final content of the presentation

Oocyte/Follicle Development

Decreasing numbers and increasing death
Why culture follicles?

- Define the fundamental mechanisms of oocyte development (basic science)
- Clinical applications: Fertility preservation – restore fertility
- Agriculture
- Genetic modification
- Endangered species
- Germ line preservation
- Toxicity testing

Culture of Preantral Follicles: Rodent Models

- Whole preantral follicles
  - Physiological model
  - Useful for study of mechanisms of oocyte/follicle development
  - Require manual dissection

- Oocyte-granulosa cell complexes
  - Maintain oocyte-granulosa cell interaction
  - Useful for production of mature oocytes
  - Easily isolated using enzymes (e.g. collagenase)
Mural Granulosa Cells
Endocrine function

Cumulus Granulosa Cells
Developmental function

**Cell-Cell-Matrix Interactions**

**Oocyte-follicle interaction**
- Oocyte growth
- Cumulus expansion
- GC proliferation/differentiation
- Steroidogenesis
- Theca formation

**Granulosa-theca interaction**
- Proliferation
- Steroidogenesis

**ECM-granulosa/theca interaction**
- Morphology
- Proliferation
- Steroidogenesis
- Survival

**Culture Systems for Mouse Preantral Follicles/Oocytes**

- Manual Dissection
- Whole follicles cultured individually in wells
- Oocyte-granulosa cell complexes cultured on Collagen-impregnated membranes

- Collagenase

Nayudu/Gosden/Spears
Smitz/Cortvrindt
Eppig
Oocyte-granulosa cell complexes isolated from 12 day old mice

Mouse Follicle Culture Systems

1955
In vitro maturation

1979
In vitro oocyte growth

1984
IVF and Development to Live Young

1989
In vitro maturation

1995
IVF and Development to Live Young

Culture systems that support development of follicles from domestic species (cattle and sheep)
DEVELOPMENT OF CULTURE SYSTEMS

Cortical Strip Culture: Primordial Initiation

Isolated Follicles: Preantral-antral transition

Antral Follicles: Oocyte Competence

Preantral Follicle Culture

Isolated Bovine Preantral Follicles
Development of a serum free culture system


Production of MMP-9 is associated with health and antral formation


Culture System: Bovine and Ovine follicles

Ovarian Cortex

Preantral Follicle (159 ± 1.7 µm)

250 µl medium in 96-well plate

Histology

Immunocytochemistry

Radioimmunoassay
Effect of IGF-1 exposure on oocyte degeneration

![Graph showing the effect of IGF-1 exposure on oocyte degeneration.](image)


Effect of activin on oocyte growth in vitro

![Graph showing the effect of activin on oocyte growth.](image)


Oocyte Morphology

![Images of oocyte morphology.](image)
Activin/FSH combination affects

- Follicle Growth
- Antral cavity formation
- Oocyte Development

Actin Distribution & Density

Control

rhAct A

Stabilisation of Cytoskeleton and oocyte-somatic cells during in vitro development: Effect of Activin and FSH

McLaughlin et al., 2010 MeR
References: