Fertilization
Animal Development

Key Concepts

- Activation
- Changes
- Juxtaposition
- Acrosome
- Syngamy
- Membrane Potential
- Cortical Reaction
- Electrophysiology

Introduction

- Sexual Reproduction
- Fertilization
- Syngamy
- Conjugation
- Parthenogenesis
**Activation**
- Inhibition
- Fertilization
- Artificial activation
- Parthenogenesis

**Physiological Changes**
- membrane potential
- increase in Ca$^{++}$
- increase in pH
- increase in permeability
- increase in viscosity
- increase in proteolytic enzymes
- start protein synthesis
- start DNA synthesis

**Morphological Changes**
- Sperm morphology
- Surface interactions
- Egg cytoplasm
Juxaposition of Gametes

- Chemotaxis
- Chemotaxins
- Species Specific (1)
- Miller (1978)
- sea urchins
- resact
- Capacitation

Acrosomal Reaction

- sea urchin
- midpiece
- nucleus
- G-actin
- acrosome
- jelly coat
- vitelline membrane
- species specific (2)
**Acrosomal Reaction**

- Membrane fusion
- Acrosome membrane
- Sperm cell membrane

**Acrosomal Reaction**

- Digestive enzymes
- Channel
- Jelly coat

**Acrosomal Reaction**

- G-actin
- Polymerize
- F-action
- Acrosomal tubule
**Acrosomal Reaction**
- gamete fusion
- membrane fusion

**Fertilization: Hermit Crab**
- sperm
- midpiece
- nucleus
- oocyte

**Fertilization: Giant Sperm**
- Frog lung Fluke (trematode)
  - small eggs: 8 µm
  - sperm head: 20-30 µm
  - sperm tail: 275 µm
  - fertilized egg: 10 µm
- Drosophila bifurcata
  - sperm: 6 cm (male=0.3 cm)
    - translates to 40 yds for a 6 ft man
  - females: store sperm in long, thin receptacle in which sperm stretch out
Fertilization: Mammals

- bindin
- acrosomal tubule
- zona pellucida
  - (vitelline membrane)
- gamete fusion

Syngamy

- male pronucleus
- centriole
- microfilaments
- mitochondria
- flagellum

Syngamy

- nuclear membrane
- decondensation
- histones
Syngamy

- nuclear membrane
- decondensation
- histones
- centriole

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Syngamy

- nuclear membrane
- decondensation
- histones
- centriole --> movement

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Syngamy

- centriole
- spindle fibers
- aster
Syngamy
- spindle fibers
- movement
- pronuclear fusion
- 2N→ zygote

Syngamy
- centrioles
- mitotic spindles
- DNA replication

Membrane Potential
- Resting Membrane Potential (-70 mv)
- ions
- Na⁺
- membrane permeability
- +20 mv
- fast block to polyspermy
Cortical Reaction

- slow block to polyspermy
- cortical granules
- golgi
- 15,000/egg

Cortical Reaction

- fusion with cell membrane
- release contents
- cell membrane
- vitelline membrane
Electrophysiology

What is the relationship between Na+ in and H+ out?

Experiment 1
-use Na+ free H₂O
-fertilize eggs

Experiment 2
-add Na+ back into previous experiment

Which is more important: Na+ in and H+ out?

Experiment 1
-add freshly fertilized eggs to a solution of amiloride and Na+

Electrophysiology

Which is more important: Na+ in and H+ out?

Experiment 1
- add ammonia to the cytoplasm of the oocyte

Conclusions

Electrophysiology

The Role of Calcium

Heilbrum
Mazia
Steinhart & Eppel
Vacquier & Payne
ionophore A23187
aequorin

General Conclusions

Summary: Electrophysiology
### Summary: Timing

<table>
<thead>
<tr>
<th>Event</th>
<th>Timing</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sperm-egg binding</td>
<td>0 sec</td>
</tr>
<tr>
<td>Minor influx of sodium ions; changes in membrane potential (fast block to polyspermy)</td>
<td>1 sec</td>
</tr>
<tr>
<td>Sperm-egg membrane fusion</td>
<td>6 sec</td>
</tr>
<tr>
<td>Liberation of Ca++ from endoplasmic reticulum</td>
<td>6 sec</td>
</tr>
<tr>
<td>Cortical reaction; Na+/H+ exchange begins</td>
<td>15-60 sec</td>
</tr>
<tr>
<td>Conversion of NAD+ to NADP+</td>
<td>60 sec</td>
</tr>
<tr>
<td>Formation of fertilization membrane complete</td>
<td>60 sec</td>
</tr>
<tr>
<td>Rise in oxygen consumption</td>
<td>60 sec</td>
</tr>
<tr>
<td>Sperm entry</td>
<td>1-2 min</td>
</tr>
<tr>
<td>Increase in intracellular pH</td>
<td>1-5 min</td>
</tr>
<tr>
<td>Sperm chromatin decondensation</td>
<td>2-12 min</td>
</tr>
<tr>
<td>Sperm nucleus migrates toward the center of the egg</td>
<td>2-12 min</td>
</tr>
<tr>
<td>Egg nucleus migrates toward the center of the egg</td>
<td>5-10 min</td>
</tr>
<tr>
<td>Increase in protein synthesis</td>
<td>5-10 min</td>
</tr>
<tr>
<td>Activation of amino acid transport systems</td>
<td>5-10 min</td>
</tr>
<tr>
<td>Fusion of egg and sperm nuclei</td>
<td>20 min</td>
</tr>
<tr>
<td>Initiation of DNA synthesis</td>
<td>20-40 min</td>
</tr>
<tr>
<td>First cleavage</td>
<td>85-90 min</td>
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