Glucocorticoids, Drug Transporters and Programming of the Fetal Brain

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Prenatal Environment: Maternal Adversity
Glucocorticoids

Transgenerational
Mechanisms

Transporters

Developing Brain:
Hippocampus
Hypothalamus

Stress
Endocrinology
Behaviour
Learning
Disease Susceptibility

• Timing of exposure
• Sex-specific
• Age-dependent
• Transgenerational
• Mechanisms
• Pituitary-adrenal development
• Maternal glucocorticoid therapy
  • neuroendocrine
• Programming mechanisms
• Fetal drug transport
  • placenta, blood-brain-barrier
Fetal HPA Axis

CRH mRNA
AVP mRNA

CRH
AVP

POMC mRNA

POMC
ACTH

Cortisol

GR
MR

Fetal Plasma Cortisol

- Sheep
- Pig
- Human
- Guinea-pig
- Horse

Fetal plasma cortisol (ng/ml)

Period before birth (days)

Glucocorticoids are a master switch in the fetal brain:

Gene transcription
Epigenetic modification
Outline

• Pituitary-adrenal development
• Maternal glucocorticoid therapy
  • neuroendocrine
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Antenatal Glucocorticoid Therapy

• ~10% of pregnant women deliver preterm (term 40wk); increasing

• Glucocorticoids decrease respiratory distress syndrome (RDS)

• Recommended treatment: single course of synthetic glucocorticoid (GC) between 24-34 weeks

• Surveys of obstetrical practice: multiple courses of GCs (~11)
Antenatal GC: HPA Function (6-10yr)

Trier Stress Test

Mean salivary cortisol (nmol/l)

- PP/GC (n=81)
- PP/nonGC (n=43)
- Controls (n=85)

Time (min)
The Guinea Pig

- Long gestation species: neuroanatomically mature young
- Well-defined brain development profile
- Haemomonochorial placentation
- Functional corpus luteum (16-day)
- Mothers provide psychosocial, minimal metabolic support
Prenatal Glucocorticoid: Guinea Pig

Maternal Glucocorticoid Treatment (1mg/kg)

Neurogenesis | Brain Growth | Myelination
---|---|---
d40 | d50 | d60

Birth (70)

Monitor Development

Endocrine Behavior
Cardiovascular Morphological

24 80
Prenatal GC Exposure: Juvenile Female

Open-Field Exposure (Day24)

[Graph showing cortisol levels over time with significance levels indicated by *** and **].

Veh N=10
Beta N=8

P<0.05

Moisiadis, Kostaki, Matthews, SGI, 2012
Prenatal GC: Adult F₁ Male Offspring

Plasma Cortisol

- Veh
- Dex

ACTH (0.5µg/kg)

Pre 5 15 30 60 90 120

Time (Mins)

Plasma Cortisol (ng/ml)

CRH mRNA
AVP mRNA

POMC mRNA

Hippocampal MR mRNA

Region

CA1/2 CA3 CA4 DG

MR mRNA (ROD)

P<0.007

Liu et al, AJP 2001
Fetal HPA Axis:

- Metabolism, growth, repair, reproduction (manage resource allocation)
- GC affect expression >10% genome
- Modified regulation in chronic diseases

-Fully programmable-
• Pituitary-adrenal development
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• Programming mechanisms
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  • placenta, blood-brain-barrier
Mechanisms: GC Programming

• Direct actions
  • Structure
  • Wiring

• Epigenetic effects
  • GR  POMC
  • MR  CRH
Epigenetic Modification

Hippocampus
Hypothalamus
Pituitary
Placenta

The 2 main components of the epigenetic code
- DNA methylation
- Histone Modification
  - Acetylation
  - Methylation
  - Phosphorylation
  - Ubiquitination
  - Sumoylation

NATURE|Vol 441|11 May 2006
Prenatal GC: Fetal Hippocampus

Maternal sGC (1mg/kg)

<table>
<thead>
<tr>
<th>Group</th>
<th>Number of Genes</th>
<th>Increased Expression</th>
<th>Decreased Expression</th>
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</thead>
<tbody>
<tr>
<td>sGC Exposure Ctrl vs sGC</td>
<td>1135</td>
<td>684</td>
<td>454</td>
</tr>
</tbody>
</table>

Crudo et al, Endocrinology, 2013
Fetal Hippocampal Methylation: Gd52

Crudo et al, Endocrinology, 2013
Fetal Hippocampal Methylation: Gd52

Development
Gd52 vs Gd65

Effect of sGC
52 vs 52Beta

Mineralocorticoid Receptor (NR3C2)

- Timing of exposure critical
- Cortisol (MR/GR) vs synthetic GC (GR)

- Multidimensional analysis

Crudo et al, Endocrinology, 2013
Outline

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Multidrug Resistance P-Glycoprotein (P-gp)

- ABC superfamily
- Tumour cells
- Normal tissues
- Placenta
- Blood-brain barrier
- Wide substrate specificity
- Glucocorticoids
- ABCB1

Nature Reviews | Cancer
• 75% women take prescription medication in pregnancy
• 10% potential teratogenic drugs
  • Glucocorticoids: 1 in 10
  • Anti-HIV: 1 in 1000
  • Anticancer: 1 in 1000
  • Antidepressants: 1 in 10
  • Anti-arrhythmia

Andrade et al 2004; Irvine et al, 2010
Placenta: P-glycoprotein (P-gp)
ABCB1 P-gp: Human Placenta

**ABCB1 mRNA**

Weeks of gestation

**P-gp Protein**

P-gp:Gß protein ratio

Sun, Kingdom, Baczyck, Lye, Matthews & Gibb, Placenta 2005
• Placental protection decreases
• Fetal BBB protection?
Fetal Protection: Blood Brain Barrier

- Fetal Brain Capillary
- Brain
- tight junction
- P-gp
- Drug
What Modulates P-gp?

- **Glucocorticoids**  Petropoulos S et al, 2010, Placenta
- **Infection/Cytokines**  Bloise E et al, 2013, PLoS One
- **Growth factors**  Baello S et al, 2013 submitted
- **Oxygen**  Lye P et al, 2013, Placenta
- **SSRIs**  Bhuiyan M et al, 2012 Reprod Sci
• Placental protection decreases
• Fetal BBB protection increases
• P-gp responsive:
  • Glucocorticoids
  • Infection-cytokines
  • Oxygen
• Novel treatment modalities
Prenatal Environment: Maternal Adversity

Glucocorticoids

Transporters

Developing Brain: Hippocampus Hypothalamus

• Timing of exposure
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• Mechanisms

Stress ↔ Behaviour
Endocrinology Learning

Disease Susceptibility
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David Phillips
Brain Microvessel Endothelial Cells

Iqbal & Matthews, Endocrinology 2011