The Placenta: Influence on Fetal Programming and Useful Afterbirth?

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Fetal Programming
(Barker Hypothesis)
(Fetal Origins of Adult Disease)
(Developmental Origins of Health and Disease)

Life in utero determines risk of development of disease in adult life
- Cardiovascular
- Diabetes (Insulin resistance/Metabolic syndrome)
- Obesity
- Stroke
- Osteoporosis
- Obstructive Airway Disease
- Cancer
- Disordered HPAA axis
- Behavioral abnormalities
Birth weight is an easily measured surrogate for exposure to an adverse intrauterine environment.
In-Utero Exposures (Stressors) that Cause
An Adverse Intrauterine Environment

Under- or Over-nutrition
Dietary composition, changes in diet
Inappropriate exposure to developmental signals e.g. glucocorticoids
Maternal stress
Medical conditions (PE, IUGR)
Hypoxia
Oxidative stress
Environmental (epigenetic) influences
Role of the Placenta

- Anchors conceptus into uterus
- Interface between mother and fetus
- Immune barrier
- Gas exchange
- Transfers/modifies nutrients
- Secretes peptide and steroid hormones to regulate maternal metabolism and fetal growth and differentiation
- Regulatory determinants
  - Blood flow
trophoblast thickness, surface area and function
expression of transporters
Fetal Programming: Key Questions re the Placenta

• Is it an active participant or innocent bystander in programming?
• If active participant, what are mechanisms?
• Does it function in isolation— the feto-placental unit?
• Can it be used in real time or retrospectively as a surrogate for fetal exposures or experiences?
## Placental Growth and Development Throughout Gestation

<table>
<thead>
<tr>
<th>Metric</th>
<th>6 weeks</th>
<th>Term</th>
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<tbody>
<tr>
<td>Fetal/Placental Weight Ratio</td>
<td>0.18</td>
<td>7.23</td>
</tr>
<tr>
<td>Villous volume occupied by vessels (%)</td>
<td>2.7</td>
<td>28.4</td>
</tr>
<tr>
<td>Trophoblast Surface area (m²)</td>
<td>0.08</td>
<td>12.5</td>
</tr>
<tr>
<td>Mean Trophoblast Thickness (µm)</td>
<td>18.9</td>
<td>4.1</td>
</tr>
<tr>
<td>Maternofetal Diffusion Distance (µm)</td>
<td>55.9</td>
<td>4.8</td>
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</table>
Critical periods during placental development

- Implantation
- Trophoblast invasion
- Establishment of blood flow to the IVS
- Placental vascular development/Branching angiogenesis
- Trophoblast differentiation and syncytium formation
- Non-Branching angiogenesis
- Exponential fetal growth

Gestation (weeks)
Determinants of Placental Nutrient Transport and Placental Function

1. Vascular (flow) dependent
   angiogenesis and vasculogenesis, utero-placental and feto-placental blood flow

2. Trophoblast (membrane) dependent
   Exchange surface area, transporter expression and activity, hormone production and metabolism – trophoblast proliferation and differentiation
Placental Vascular and Trophoblast Development and Fetal Programming

• Fetal growth in Yorkshire pig from mid to late gestation is associated with an increasing area of endometrial attachment i.e a larger exchange area.

• Growth of Chinese Meishan fetus at this time is associated with an increase in the density of placental blood vessels leading to greater placental efficiency (fetal/placental weight) in the Meishan (Vonnahme and Ford 2004).
Placental Vascular Impedance and Fetal Heart Development

- Changes in placental vascular impedance impact fetal cardiovascular loading
- Heart fitness is determined by hemodynamic, growth factor and oxygen/nutrient cues before birth.
- All are influenced if not regulated by the placenta
Effect of Time of “Insult” on Expression of Glucose and Amino Acid Transporters

- IDDM plus LGA gives increased Glut 1 in BM and increased system A aa transporter
- GD plus LGA no change in Glut 1 in BM but increased system A
  (Consistent with hyperglycemia causing increased Glut 1 in the 1st trimester)
- System A increased in 3rd trimester not 1st trimester (Jansson)
- Glucocorticoids decrease expression and function of Glut transporters (Hahn)
Imprinted Genes and the Placenta

- All imprinted genes found in placenta where they affect growth of placental cell types including labyrinthine and spongiformrophoblast

- Paternally expressed imprinted genes enhance fetal growth
  Knockout of paternally expressed genes eg *Igf2* reduces placental growth

- Maternally imprinted genes suppress fetal growth
  Knockout of maternally expressed genes e.g. *p57kip2* results in placental hyperplasia

- Imprinted genes control fetal growth and placental growth and therefore may control the supply (placenta) and demand (fetus) for nutrients
Glucocorticoid Action and Metabolism

- $11\beta$HSD-2 in syncytiotrophoblast converts cortisol to cortisone (blocks exposure to maternal cortisol)
- $11\beta$HSD-2 increases with gestational age
- $11\beta$HSD-2 increases at time of oxygen switch (hypoxia decreases $11\beta$HSD-2)
- Mutations in $11\beta$HSD-2 give IUGR
- Decreased $11\beta$HSD-2 with hypoxemia and PE
- Nutritional restriction causes decreased $11\beta$HSD-2
Preeclampsia and IUGR

- Characterized by defective trophoblast invasion
- Ischemia/reperfusion injury, hypoxia
- Inflammatory response
- Oxidative stress
- Nitrative stress
- Functional effect on placenta
Oxidative Stress and the Placenta

Pregnancy per se is a state of oxidative stress
Increased further in preeclampsia, IUGR and diabetes
Increased morbidity and mortality
Endothelial and vascular dysfunction
Increased apoptosis, ER stress, UPR response
Increased deportation of trophoblast
Increased hsp70 and nitrotyrosine
Obesity has become a major health problem in the US. In women of reproductive age: 56.7% are overweight (BMI >25) 30.2% are obese (BMI >30)*.

Obesity during pregnancy is linked to maternal complications and poor perinatal outcome PIH, Diabetes, Increased caesarean delivery and complications, Prematurity, Stillbirth, Macrosomia

As adults the offspring show increased incidence of: obesity insulin resistance hypertension cardiovascular disease.

Obesity is an inflammatory condition.
Inflammation

Superoxide/Nitric oxide

Oxidative/Nitrative Stress

Altered Placental Function

Altered Maternal/Fetal Metabolism

Pregnancy Outcome
$O_2$ + NADPH oxidase + Xanthine oxidase → $O_2^-$ + NO. → ONOO$^-$

L-arginine + $O_2$ → NOS → NO.

$O_2^-$ + SOD → $H_2O_2$

Oxidative stress

Nitrative stress (protein nitration)
Western Blot Analysis of Placental Homogenates

Intensity (Nox/vimentin)

Nox1
Nox5
vimentin
vimentin

p=0.034
p=0.001
Reactivity of the Fetal-Placental Vasculature to U46619

Change in Perfusion Pressure (mmHg)

<table>
<thead>
<tr>
<th></th>
<th>control</th>
<th>diabetic</th>
<th>preeclamptic</th>
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<tbody>
<tr>
<td>n=6</td>
<td></td>
<td>n=6</td>
<td>n=5</td>
</tr>
<tr>
<td>-9 -8 -7</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>p&lt;0.01</td>
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U46619 (M)
P38 MAP Kinase and the Placenta

Regulates the development and differentiation of various cell types

p38MAPK -/- null mouse is embryonic lethal (loss of labyrinth and reduced spongiotrophoblast layers, reduced vascularisation of labyrinth and increased apoptosis)

Lack of p38 causes inadequate trophoblast invasion

Regulates placental lactogen gene in trophoblast
Phospho p38 In Nitrotyrosine IP

Equal amounts of protein were immunoprecipitated with anti-nitrotyrosine antibody and western blotted with phospho-p38 antibody.
Phospho-p38 MAPK Activity In Phospho p38 Immunoprecipitates

NORMAL

PREECLAMPTIC

p-ATF2

Band intensity (relative units)

Fig. 11

p<0.05
Hypoxia

Inflammation

Maternal substrates

MATERNAL CIRCULATION

NO + O2- → ONOO

Protein Nitration

PLACENTA

FETUS

Ligand

Receptor

Membrane Transporter e.g. GLUT, amino acid

Maternal substrates

Structural Proteins

Signal Transduction Molecules

Enzymes

Cellular Metabolism

Fetal growth/differentiation signals

Fetal Substrates

Peptide/Steroid Hormones

Production/Metabolism
Can we use the placenta as a diary of fetal exposures??
What can be measured in utero?

• Blood flows and resistance to flow.
• Peptide production, used as indices of
  – Trophoblast invasion
  – Angiogenesis/antiangiogenesis
  – Regulation of metabolism
• Steroid secretion
• Inflammatory state
What can be measured post-delivery?

- Weight, Size, Shape, Surface area
- Histologic parameters, vascular and trophoblast
- Vascular reactivity and compliance
- Metabolic rate
- Function
  - transport,
  - Peptide synthesis,
  - steroid synthesis
- Content of metabolites, xenobiotics, metals
- Redox state
- Epigenome
Caution!

- Collection – C section
- Random sampling protocol
- Gestational age
- Medical conditions
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Oxygen and Development of Villous Vascular Tree and Trophoblast Differentiation
## Causes and Consequences of Altered (Adaptive) Placental Function

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<th>Cause</th>
<th>Consequence</th>
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<tbody>
<tr>
<td>Hypoxia</td>
<td>Early pregnancy loss</td>
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<tr>
<td>Oxidative/nitrative stress</td>
<td>Abnormal vascular development</td>
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<td></td>
<td>Altered trophoblast development</td>
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<td></td>
<td>Altered transporter activity</td>
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<td>Altered steroidogenesis</td>
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<tr>
<td>Altered vascular development</td>
<td>Abnormal blood flows and resistance</td>
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<td></td>
<td>Altered exchange surface area</td>
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<td></td>
<td>Altered substrate supply</td>
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<tr>
<td>Altered trophoblast differentiation</td>
<td>Altered substrate metabolism and steroidogenesis</td>
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<td></td>
<td>Altered transporter function</td>
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<td></td>
<td>Altered barrier thickness</td>
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<tr>
<td>Altered steroidogenesis</td>
<td>Altered/abnormal signals to mother and fetus</td>
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