Role of Progesterone Receptor Dysregulation in the Pathogenesis of Preterm Birth: Why would Progesterone supplementation work?
Does Progesterone (P4) prevent spontaneous PTB?

Meis et al (N Engl J Med. 2003) randomized 459 women with prior spontaneous singleton PTB <37 weeks to weekly 250 mg 17-OHP (i.m.) vs. placebo from 16-20 weeks until 36 weeks and noted 17-OHP reduced risk of delivery:

- <37 weeks (36 vs 55%) [RR 0.66; 95% CI: 0.54-0.81]
- <32 weeks (11 vs 20%) [RR 0.58; 95% CI: 0.37-0.91]

Dodd et al conducted a meta-analysis (Cochrane Syst Rev. 2013) of RCT of P4 administration for prevention of recurrent PTB (11 trials, n = 1899 women) and noted P4 lowered risks for:

- Birth <34 weeks [RR 0.31, 95% CI: 0.14-0.69]
- Birth <37 weeks [RR 0.55, 95% CI: 0.42-0.74]
- Neonatal death [RR 0.45, 95% CI: 0.27-0.76]
Does P4 prevent spontaneous PTB?

OPPTIMUM trial (Lancet. 2016) 1228 women at high risk (prior PTB ≤34 weeks, CL ≤25 mm, or positive fFN, plus clinical risk factors) randomized to vaginal P4 200 mg daily or placebo from 22-24 weeks to 34 weeks. Major findings were that P4 did not significantly reduce fetal death or PTB < 34 weeks or severe neonatal outcomes.

PROGRESS trial (Plos Med. 2017) randomized 787 women with prior PTB to vaginal P4 100 mg vs. placebo from 20-24 weeks to 34 weeks and observed no difference in PTB rates, birth weights or prevalence of RDS or serious neonatal sequelae.
Central Role Played by Decidua in Mediating Parturition

**Physiological:** Source of PG, IL-8, IL-6, and MMPs.

**Pathological:** Source of IL-1β and thrombin in infection and abruption associated PTB, respectively.
Cell Types at the Maternal-Fetal Interface

T : Trophoblast
NK : Natural Killer Cell
DC : Decidual Cell
M : Macrophage
L : T-lymphocyte

*** Of note that decidual cells are the major cell type (>50%) at the maternal-fetal interface
PR Expression is restricted to Decidual Cells at the Maternal-Fetal Interface

RED: Vimentin (identifies decidual cells)
Gray: Cytokeratin (identifies Interstitial trophoblast)
Brown: PR or GR (arrows for DCs; arrowheads for IT’s)
PR Expression in Decidua Parietalis and Basalis

Parietalis/Pre-Contract

Basalis/Pre-Contract.

Parietalis/Contraction

Basalis/Contraction

Brown: PR Red: Vimentin

DC: arrows IT: arrowheads

(P<0.05)

(P<0.05)

(Lockwood et al. JCEM 2010;95(5):2271-75)
SUMMARY-I

- PR expression is restricted to decidual cells at the maternal-fetal interface.
- Decidual cell PR expression significantly decreases in laboring vs. non-laboring specimens at term.
- GR expressed primarily by interstitial trophoblast and its expression does not change in laboring vs. non-laboring specimens.
Decidual-Chronic Inflammation and PTB

- BV is associated with PTB (OR 1.95-2.19) and facilitates over-growth of mycoplasma sp., gardnerella, gram (-) bacteria, strep. and staph (Am J Ob Gyn 1995; 173:1231-5; NEJM 1995;333:1737-42; Ob/Gyn 1996;87:656-60).

- Prevalence of a Lactobacillus-poor vaginal community state type (CST 4) inversely correlated with GAD (P = 0.0039). PTD Risk greater with CST 4 plus Gardnerella or Ureaplasma. (Proc Natl Acad Sci U S A. 2015 Sep 1;112(35):11060-5.)
Decidual-Chronic Inflammation and PTB

- Bacterial products (endotoxins and exotoxins) bind to decidual and fetal membrane amnio-chorion TLR-4 and -2 to induce local IL-1β \((\text{AJOG} 2003; 189:139-47; \text{Infect Immun} 2004; 72:5799-806; \text{AJOG} 2004; 191:1346-55; \text{Immunology}. 2002; 107:145-51)\).

- IL-1β enhances levels of NF-κB in decidua and fetal membranes to induce MMP-1,3, & 9, and COX-2 expression while inhibiting chorion PGDH \((\text{Mol Human Reprod} 2001; 7:81-6)\).

- Of 1025 women with spontaneous PTB < 34 weeks, chorioamnionitis/IUI was present in 38% \((\text{Am J Obstet Gynecol} 2015; \text{Feb. pii: S0002-9378(15)00132-5. E-pub})\).
Decreased Nuclear PR Expression in DCs at the Maternal-Fetal Interface of CAM-associated PTB

Cultured Human Primary Term Decidual Cells
IL-1β Reduces PR mRNA and Protein Expression in Cultured Term DCs.

* $p < 0.05$ vs. $E_2$; # $p < 0.05$ vs. $E_2$

$\psi$ $p < 0.05$ vs. $E_2 + MPA$

IL-1β Induces COX-2, but not Cox-1 Expression in Cultured Term DCs

*p < 0.05 vs. E2;
#p < 0.05 vs. E2 + MPA;
ѱ p < 0.05 vs. E2+IL-1β.

IL-1β Induces Secretion of PGE$_{2}$ and PGF$_{2α}$ in Term DC Cultures

**PGE$_2$ ELISA in CMS.**

* $p < 0.05$ vs. $E_2$
# $p < 0.05$ vs. $E_2 +$ MPA
Ψ $p < 0.05$ vs. $E_2 +$ IL-1β

**PGF$_{2α}$ ELISA in CMS.**

* $p < 0.05$ vs. $E_2$
# $p < 0.05$ vs. $E_2 +$ MPA
Ψ $p < 0.05$ vs. $E_2 +$ IL-1β

PGF$_{2\alpha}$ but not PGE$_2$ Reduces PR Expression in Term DC Cultures

Immunoblot analysis of PR levels in term DCs treated with E$_2$ + MPA or Org ±10$^{-6}$ M PGE$_2$ or PGF$_{2\alpha}$ for 24 h.

Org: Org2058 (pure progestin)

Note that PGE$_2$ did not change whereas PGF$_{2\alpha}$ significantly reduced DC PR levels.

Inhibition of COX-2 does not Reverse IL-1β Suppressed PR Expression in Term DC Cultures

**Indo**: Indomethacin, (10⁻⁶ M, non-selective COX inhibitor)

**Of note**: Indomethacin alone has no effect on PR levels, while its combination with IL-1β induces further decreases in PR levels.

IL-1β Activates

Multiple Signaling Cascades in Term DCs

Immunoblot analysis of following 15 min E₂ + MPA ± IL-1β treatment of term DCs for phospho (Ph-) and total (T-) levels of:

1) p65 NF-κB,
2) ERK1/2 MPAK,
3) p38 MAPK,
4) AKT
Inhibition of ERK1/2 MAPK Reverses IL-1β Reduced DC expressed PR

Immunoblotting of term DCs treated with: E₂+MPA (EM) ± IL1β ± NF-κB inhibitor III (NFIII) or ± ERK1/2 inhibitor PD98059 (PD) or ± p38 MAPK inhibitor SB203580 (SB) for PR-A and PR-B expression.

Note that ERK1/2 inhibitor (PD) significantly reverses IL-1β reduced PR levels, whereas neither the NF-κB inhibitor (NFκB III) nor the p38 inhibitor (SB) reversed this effect.

Expression of Ph-ERK1/2 and T-ERK1/2 MAPK in DCs and ITs in GA-matched Control vs. Abruptio-Related Decidua

DCs: Arrowheads
ITs: Arrows

SUMMARY-II

- Decreased PR expression in CAM vs. GA-matched control.
- IL-1β inhibits PR mRNA and protein levels in cultured decidual cells.
- This inhibitory effect on PR expression is primarily mediated by IL-1β activation of phospho-ERK1/2 signaling.
STRESS AND PTB

Combination of Depression and PTSD strongly linked to PTB (OR of 4.08; 95% CI: 1.27-13.15).

SSRI, both with (OR= 2.1; 95% CI: 1.0-4.6) and without (1.6; 1.0-2.5) MDD is associated with PTB

FKBP51 expression increased in frontal cortex of MDD patients and SNPs associated with FKBP51 over-expression linked to MDD/PTSD

FKBP-51/52 Interaction with the Steroid Receptors

FKBP51 or FKBP52

Steroid Receptor (PR, GR etc.)
FKBP51 and 52

- FKBP51 and 52 differentially affect Steroid Receptor functions.
- FKBP52 potentiates PR and GR activity.
- FKBP51 attenuates PR and GR responses.
- FKBP51 and 52 assemble with ER, but neither co-chaperone affects ER activity.
Expression of FKBP51 at the Maternal-Fetal Interface

Before Contraction

After Contractions

Decidua basalis

Decidua parietalis

FKBP51: Brown
Vimentin: Red
Cytokeratin: Grey
DC: Arrows
ITs: Arrowheads

FKBP51 mRNA Levels in Term DC, Cytotrophoblast and Syncytiotrophoblast Cultures

FKBP51 mRNA expression following 6 h of treatment with dexamethasone (Dex).

TDC: Term decidual cells;
CTY: Cytotrophoblast;
SCT: Syncytiotrophoblast

FKBP51 and FKBP52 mRNA Expression in Term DCs

FKBP51 and 52 mRNA expression following 6 h of treatment of Decidual cells with: E₂; MPA, a mixed progestin-glucocorticoid; Dex, a pure glucocorticoid; and Org2058 (Org), a pure progestin.

FKBP51 reduces PR binding to DNA (PRE) by EMSA

Role of FKBP51 in Stress-Induced PTB using FKBP51-/- Mice

FKBP51 WT ♂ X FKBP51 WT ♀  
FKBP51 WT ♂ X FKBP51 KO ♀

♂ 12-14 weeks old ♀ 6-8 weeks old  
♂ 12-14 weeks old ♀ 6-8 weeks old  

Mating x 4h  
Vaginal smear/sperm+ 0 d.c.

Restrain stress was applied 1h x 3 each day on days E8 to E18  
Morning serum collected within 30 min of 1st stress treatment on E11  
Between day E18-22, delivery controlled at each 4 h interval.
Prolong Gestation in Non-Stressed 
FKBP51^{-/-} vs FKBP51^{+/+} Mice

<table>
<thead>
<tr>
<th>Gestational Length</th>
<th>Non-Stress</th>
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</thead>
<tbody>
<tr>
<td>FKBP51^{+/+}</td>
<td>19.10 ± 0.26 (N=10)</td>
</tr>
<tr>
<td>FKBP51^{-/-}</td>
<td>20.25 ± 0.29 (N=11)</td>
</tr>
</tbody>
</table>

* p=0.01 KO NS vs WT NS

Mean ± SE: 20.25 ± 0.29 (N=11) vs. 19.10 ± 0.26 (N=10)
Prenatal Stress induces PTB in WT Mice

Mean ± SEM; 18.72 ± 0.03 (N=10) vs. 19.10 ± 0.26 (N=10)

* p=0.018 WT ST vs WT NS

FKBP51^{+/+}
Gestational Length is Significantly Longer in Stressed induced FKBP51 KO vs. WT mice

* p=0.002 KO ST vs WT ST

Mean ± SE; 19.45 ± 0.21 (N=10) vs. 18.72± 0.03 (N=10)
Similar Maternal Body Weight Gain and Food Intake during Pregnancy in Stressed and Non-stressed Mice
SUMMARY - III

- FKBP51 is mainly expressed by DCs vs ITs and that decidual FKBP51 expression increases in labor.
- FKBP51 KO mice have longer gestational length and are more resistant to stress-induced PTB.
- FKBP51 KO female mated with KO male mice are more resistant to stress-induced PTB compared to WT or KO females mated with WT male.
Abruption

- ↑ Thrombin PAR

Chorioamnionitis

- ↑ IL-1β, TNF-α

- ERK1/2 MAPK

- COX-2
- ↑ PGF2α
- ↓ PR Levels
- ↓ PR Activity
- ↑ MMP-3

Stress

- ↑ GR signaling

- FKBP51

- Membrane rupture and Cervical ripening

PTB

- ↑ Contractions
Role of Progesterone Receptor Dysregulation in the Pathogenesis of Preterm Birth: Why would Progesterone supplementation work?