

9 **PREECLAMPSIA IS CHARACTERIZED BY A SOLUBLE SELECTIN PROFILE CONSISTENT WITH LEUKOCYTE AND ENDOTHELIAL CELL ACTIVATION** TINNAKORN CHAIWORAPONGSA¹, JU CHEOL KIM¹, YEON MEE KIM¹, GIANCARLO CONOSCENTI¹, KYO HOON PARK¹, SAM EDWIN¹, BO HYUN YOON², ROBERTO ROMERO¹; ¹Perinatology Research Branch, Detroit, MI; ²Seoul National University, Seoul

OBJECTIVE: An exaggerated activation of the innate limb of the immune response has been implicated as the cause of the maternal syndrome of preeclampsia. Intravascular inflammation would lead to endothelial cell dysfunction and activation. Selectins are a family of three separate but closely related proteins that mediate adhesion of leukocytes to endothelial cells and, thus, have been implicated in the pathophysiology of preeclampsia. L-selectin is expressed on leukocytes, P-selectin within the storage granules of endothelial and platelets and E-selectin in cytokine activated endothelium.

STUDY DESIGN: A cross-sectional study was conducted to determine the concentrations of the soluble forms of L-, E- and P-selectins in non-pregnant women (n = 20), normal pregnant women (n = 22) and patients with preeclampsia (n = 22). Patients with preeclampsia were matched for gestational age with normal pregnant women. Concentrations of selectins were determined with sensitive and specific immunoassays. Non-parametric statistics were used for analysis.

RESULTS: 1) Normal pregnancy was associated with a significant increase in the maternal plasma concentration of sP-selectin, a decrease in sL-selectin and no changes in sE-selectin (see Table); 2) Preeclampsia was associated with a significant increase in sP-selectin, sE-selectin and a decrease in sL-selectin (see Table).

CONCLUSION: 1) Normal pregnancy is associated with a selectin profile consistent with evidence of intravascular leukocyte activation; 2) Preeclampsia is characterized by both leukocyte activation and cytokine-induced endothelial cell activation.

Table

	SP-SELECTIN	SE-SELECTIN	SL-SELECTIN
Non-pregnant women (n = 20)	56.5	38.6	964.0
Normal pregnant women (n = 22)	91.3*	42.9	730.5*
Preeclampsia (n = 22)	103.4**	59.9**	574.8**

Value expressed as median (ng/ml); *P < .05 compared to non-pregnant women; **P < .05 compared to normal pregnant women.

10 **EFFECT OF PREGNANCY ON VASCULAR REACTIVITY IN TRANSGENIC MICE LACKING A FUNCTIONAL ENDOTHELIAL OR INDUCIBLE NITRIC OXIDE SYNTHASE** MONICA LONGO¹, VENU JAIN¹, YURI VED-ERNIKOV², GEORGE SAADE¹, ROBERT GARFIELD¹; ¹University of Texas Medical Branch, Obstetrics and Gynecology, Galveston, TX; ²University of Texas Medical Branch, Obstetrics & Gynecology, Galveston, TX

OBJECTIVE: To compare the effect of pregnancy on in-vitro vascular reactivity in mice lacking the expression of endothelial NOS (eNOS-KO) and inducible NOS (iNOS-KO).

STUDY DESIGN: Female eNOS-KO (C57BL/6J-NOS3^{-/-}) and iNOS-KO mice (B6/129FJNOS2^{-/-}) obtained from Jackson Laboratory were used (n = 8-10/group). Two millimeter segments of aorta from nonpregnant, mid-pregnant (day 14) and term-pregnant (day 19) mice were mounted in a wire myograph for isometric tension measurements and concentration-response relationships to representative contractile and relaxing agents were determined. Data was analyzed using Student's *t* test or one-way ANOVA. P < .05 was considered statistically significant.

RESULTS: Phenylephrine (PE) contraction was decreased in nonpregnant iNOS-KO compared to eNOS-KO (Maximal effect: 28.6 ± 10.6 versus 146.5 ± 15.2; P < .05). PE responses were not different between the KO mice at midpregnancy or term. In non-pregnant, mid-pregnant, and term-pregnant mice, relaxation to acetylcholine was absent in eNOS-KO and present in iNOS-KO. Relaxation to sodium nitroprusside was not different between iNOS-KO and eNOS-KO in the nonpregnant and term pregnant, but the responses were significantly greater in eNOS-KO compared to iNOS-KO at midpregnancy.

CONCLUSION: NO is the predominant endothelial relaxing factor in pregnant mouse aorta. Pregnancy induces compensatory changes in vascular reactivity in response to genetic defects in the enzymes producing NO, especially the endothelial type. The presence of parallel compensatory mechanisms specific to pregnancy may be a teleological development aimed at the preservation of the species. Disturbances in multiple mechanisms are therefore needed in order to result in abnormal vascular function in pregnancy, preeclampsia being the primary example.

11 **MATRIX-METALLO-PROTEINASES AND LEUKEMIA-INHIBITORY-RECEPTOR ARE CO-EXPRESSED IN PERIARTERIAL EXTRAVILLOUS TROPHOBLAST IN PREECLAMPSIA** FRANK REISTER¹, JOHN KING-DOM², HANS-GEORG FRANK³, WERNER RATH⁴, PETER KAUFMANN³, BERTHOLD HUPPERTZ³; ¹Mount-Sinai Hospital, Toronto, Ontario; ²Mount Sinai Hospital, Toronto, Ontario; ³University of Aachen, Anatomy, Aachen; ⁴University of Aachen, Obstetrics and Gynecology, Aachen

OBJECTIVE: Interactions between leukemia inhibitory factor (LIF) and its receptor (LIF-R) in trophoblast cells is known to induce secretion of oncofetal fibronectin (oFN) and to modulate the expression of Matrix-Metallo-Proteinases (MMP). Therefore we speculated that an increased expression of LIF-R by periarthelial trophoblast cells in Preeclampsia (PE) could decrease their invasiveness by induction of an anchoring extravillous phenotype and by impaired expression of matrix-degrading MMP.

STUDY DESIGN: Placental bed biopsies of patients with severe PE and healthy pregnant women (n = 8 each) were immunohistochemically screened for trophoblast cells (cytokeratin) and their expression of MMP-3, MMP-7 and LIF-R. Immunostaining for CD16 detected uterine Natural-Killer (uNK) Cells.

RESULTS: Trophoblast cells accumulating in intimate neighbourhood to non-invaded spiral arteries could be detected only in specimen of preeclamptic patients. These cells showed absent expression of MMP-3 in 86% and of MMP-7 in 90%. Virtually all of these cells strongly expressed LIF-R. In healthy pregnancies the majority of interstitial as well as endovascular trophoblast cells strongly expressed MMP-3 and MMP-7, whereas reactivity for LIF-R was seen only in 25% (20-40%) of interstitial and not at all in endovascular trophoblast cells. The concentration of uNK-Cells was significantly increased in the placental bed of preeclamptic women.

CONCLUSION: LIF is a product of uNK-Cells which are increased in the placental bed of PE-patients. LIF seems to play several roles in the pathophysiology of disturbed trophoblast invasion in PE. Increased secretion of oFN affects the motility of trophoblast cells and therefore their invasiveness. Impaired secretion of fibronectin-degrading enzymes further on alters their motility. Besides, as LIF-LIF-R interaction is known to induce apoptosis in several tumor cell lines, this may contribute to apoptosis of periarthelial trophoblast in PE.

12 **DIFFERENTIAL QUANTIFICATION OF PLACENTAL APOPTOSIS AMONG NORMAL, MILD PREECLAMPSIA, AND SEVERE PREECLAMPSIA WITH OLIGOHYDRAMNIOS IN HUMAN PREGNANCY** ROBERT ORGAN¹, LAURA MARTIN², LISA PIERCE³, JAMES WRIGHT³, TROY PATIENCE⁴, BYRON CALHOUN⁵, RODERICK HUME JR⁴; ¹Madigan Army Medical Center, Developmental Pediatrics, Ft. Lewis, WA; ²MAMC, Genetics, Ft. Lewis, WA; ³MAMC, Clinical Investigation, Ft. Lewis, WA; ⁴Madigan Army Medical Center, Clinical Investigation, Ft. Lewis, WA; ⁵Madigan Army Medical Center, Maternal Fetal Medicine, Ft. Lewis, WA

OBJECTIVE: Increased placental apoptosis has been reported in pregnancies complicated by preeclampsia, and intrauterine growth retardation. Preeclampsia, prematurity, oligohydramnios and birth asphyxia have been implicated in perinatal neurologic injury. Does apoptotic index predict severity of maternal disease or neonatal outcome?

STUDY DESIGN: Placental tissue was anonymously collected from normal control and preeclamptic pregnancies. Coded tissue blocks were submitted for fixation and thin section. The PI blinded to clinical status used the method modified from Gevrieli for Terminal Deoxynucleotidyl Transferase (TdT)-mediated dUTP nick end labeling (TUNEL) to label apoptotic placental nuclei. Apoptotic indices were compared among cohorts.

RESULTS: Complicated preeclampsia with oligohydramnios demonstrated significantly increased apoptotic index (19.7 ± 9.8, P = .0004) than either preeclampsia (8.8 ± 4.9) or normal control (5.5 ± 3.0).

CONCLUSION: The apoptotic index predicts severity of maternal disease. Previous work from our lab has shown differential gene expression for cell cycle regulators in pregnancies complicated by oligohydramnios. The apoptotic index may be an effective predictor of perinatal neurologic susceptibility (cerebral palsy) measuring the phenotypic expression of accelerated placental senescence related to severity of maternal disease.