

280. Prostaglandin E₂ (PGE₂) Induces Human Endometrial Fibroblast Cell Decidualization In Vitro via PGE₂ Receptor 2 (PTGER2).

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Prostaglandins, especially of the E series, play an important role in endometrial fibroblast differentiation into decidual cells (decidualization) during embryo implantation in many species. To study the regulation of human decidualization by PGE₂, we utilized the human uterine fibroblast (HuF) in vitro cell model. Treatment of HuF cells with PGE₂ (10 microM) alone for 1-3 days caused significant (P<0.01) increases in the expression of the decidual markers *IGFBP1*, *PRL* and *FOXO1A* compared to cells treated with vehicle. This increase was significantly (P<0.01) greater in the presence of the steroids, progesterone and estradiol. Since induction of decidualization can be also stimulated by 8Br-cAMP, we hypothesized that PTGER2 and/or PTGER4 were, at least in part, responsible for mediating PGE₂-induced decidualization. To test this, we incubated the cells with 17-PT-PGE₂ (10 microM), butaprost (10 microM) or Cay10580 (1 microM) which are PTGER1, 2 and 4 agonists, respectively. In the presence of steroids, exposure to 17-PT-PGE₂ or Cay10580 for 2 days induced a small but significant (P<0.05) increased expression of *IGFBP1* and *PRL*. However, the PTGER2 agonist butaprost dramatically increased (P<0.001) the expression of these genes. To confirm these findings, we incubated the cells with the PTGER 1,2 and 4 competitive antagonists SC19220, PF04418948 or L-161,982 (10 microM), respectively. In the presence of steroids, SC19220 and L-161,982 only slightly (P<0.05) decreased PGE₂-induced (1 microM) *IGFBP1* and *PRL* expression. However, PF04418948 greatly (P<0.001) suppressed PGE₂-induced *IGFBP1* and *PRL* expression suggesting PTGER2 is principally responsible for mediating PGE₂-induced decidualization of HuF cells. Finally, we monitored expression levels of *PTGER1*, *2* and *4* receptors in HuF cells to determine whether they change during 8Br-cAMP induced decidualization. Interestingly, HuF cells express all three receptors initially but expression of *PTGER1* and *PTGER4* dramatically decreased during decidualization while that of *PTGER2* significantly increased. In conclusion, the results of this study suggest that PGE₂-induced decidualization of HuF cells is mediated mainly via the PTGER2 receptor. (This work was supported by Research Seed Grant from SIU School of Medicine to BB, SIU CURCA Undergraduate Student Assistantship to DD, and R15HD073868 to DT).

281. Endometrial Expression of Genes Involved in Growth Factor, Cytokine, Hormone, and WNT Signaling During the Early Estrous Cycle of the Cow.

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Knowledge of the molecules used by the maternal reproductive tract to regulate development of the preimplantation embryo is largely incomplete. To identify possible candidates for this function, an experiment was conducted to assess expression patterns during the first seven days after ovulation for 93 genes that could potentially be involved in control of development. Included were genes for 29 growth factors, 11 cytokines, 22 interleukins, 3 hormones, 19 WNT ligands and 9 WNT regulatory molecules. Ovulation was synchronized in 15 cows and cows were slaughtered at days 0, 3, 5 and 7 relative to predicted ovulation. Reproductive tracts were obtained and intercaruncular regions of endometrial tissue were harvested for gene expression analysis from uterine horns ipsi- and contralateral to the corpus luteum. Abundance of specific mRNA molecules was determined using the NanoString nCounter analysis system. Data were normalized against 6 housekeeping genes (*ACTB*, *ERK1*, *GAPDH*, *RPL19*, *SLC30A6*, *SUZ12*) and internal positive controls. Genes were considered expressed if the number of reads was greater than two standard deviations above the mean of negative controls. Data were analyzed by analysis of variance using the GLM procedure of SAS with day, side, and day*side as fixed effects, and cow as random effect. All genes were detected at one or more days. There was wide variation in the magnitude of gene expression. Among the 20 most expressed genes, the reads varied from 180 to over 33700. For day 5, the ten most highly expressed genes in descending magnitude were *WNT5A*, *CXCL3*, *TDGF1*, *CTGF*, *SFRP1*, *GRO1*, *IK*, *HDGF*, *VEGFA*, *IGF1*. For day 7, the ten most highly expressed genes were *TDGF1*, *WNT5A*, *CTGF*, *VEGFB*, *IK*, *HDGF*, *SFRP1*, *VEGFA*, *HDGFRP2*, *IGF2*. Nineteen genes were significantly affected by day with values highest at estrus (*BMP7*), Day 3 (*CXCL10*, *EGF*, *IL18*), Day 5 (*CSF2*, *CX3CL1*, *FGF2*, *GRO1*, *IL1A*, *IL1B*, *IL6*, *IL8*, *SFRP4*, *TGFB1*, *WNT4*) or Day 7 (*FGF12*, *HDGF*, *PGF*, *WNT9A*). Expression was higher for endometrium ipsilateral to side of ovulation for 6 genes (*CXCL10*, *FGF2*, *HDGFRP3*, *SFRP4*, *WNT4*, *WNT7A*); while expression was higher on the contralateral side for 16 genes (*CCL14*, *CSF2*, *CX3CL1*, *FGF1*, *FGF12*, *GRO1*, *IL1A*, *IL1B*, *IL8*, *TGFB1*, *VEGFB*, *WIF1*, *WNT2B*, *WNT5A*, *WNT9A*, *WNT11*). There was a significant effect of interaction between day and side for 13 genes (*DKK1*, *DKK3*, *IL12B*, *IL16*, *IL34*, *INHBA*, *NGF*, *SFRP1*, *TGFB3*, *WNT16*, *WNT2*, *WNT5B*, *WNT6*). This experiment points to potential maternal regulators of embryonic development. Further studies are needed to determine the effect of these maternally secreted molecules in embryonic development to classify them as embryokines. (Support NIH HD080855).

Poster Topic Area 2: Epigenetics of Reproduction, Nutrition and Gamete/Embryo Development, & Effect of Environment on Ovary

EPIGENETICS OF REPRODUCTION: Program Numbers 282-314

282. The Analysis of Telomere Length and Telomerase Activity in Cloned Korean Native Beef Cattle (Hanwoo).

Hyun Kim¹, Moo Y. Cho¹, Seongsoo Hwang², Changyong Choe³, Byoung-Chul Yang⁴, Yeoung-Gyu Ko³, Hwan-Hoo Seong³.

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News and updates about the SSR 2015 Annual Meeting in San Juan, Puerto

21 June 2015

Fun Run Results

Congratulations to everyone who participated in the SSR 2015 Fun Run!

Top Male Finishers

- | | |
|------------------|-------|
| 1. Lukas Chmatal | 20:20 |
| 2. Dieter Egli | 20:52 |
| 3. Ryan Matzuk | 22:39 |

Top Female Finishers

- | | |
|--------------------|-------|
| 1. Bluma Lesch | 23:27 |
| 2. Pavla Dostalova | 25:11 |
| 3. Sarah McCloski | 25:14 |