Estradiol-Induced Labor Initiation: Changes on Gene Expression of Maternal Uterine and Cervical Tissues in Periparturient Ewes.

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The onset of labor is a complex process involving various hormonal and physiological pathways that remain to be completely understood. Herein, we investigated the influence of estradiol (E2) in the initiation of parturition of periparturient Rambouillet ewes. To this end, two experiments evaluated the relationship between E2 circulating levels, birth timing (Exp.1), and gene expression (Exp.2) on the myometrium, endometrium, cervix, and caruncle tissues. Both experiments utilized the same approach but with varying E2 dosages. Multiparous ewes between 139 to 142 days of gestation were randomly assigned to either the E2 group, which received six Silastic implants of 50 mg of E2 in Exp.1 (300 mg/ewe; n=12) and four in Exp.2 (200 mg/ewe; n=6); or the control (C) group, which received 6 Silastic empty implants in Exp.1 (n=12) and 4 in Exp.2 (n=6). The implants were inserted subcutaneously in the axillary region of the ewe and removed two days after parturition (Exp.1) or at the time of slaughter, 26h after treatment (Exp.2). Maternal blood samples were drawn one day before treatment initiation in Exp.2 and 8h (Exp.1) or 26h (Exp.2) from the beginning of treatment to measure circulating levels of E2 and progesterone (P4) using immunoassay. Cross sections of the uterus, caruncles, and cervix were collected for RNA-Seq analysis. After RNA-Seq data quality control and mapping, differentially expressed genes (DEGs) were identified using DESeq2. Results indicated that before treatment, E2 levels between groups were similar. However, at 8h (Exp.1) and 26h (Exp.2) after treatment, E2 concentration was significantly higher in the E2 group (192.91±17.32 vs. 10.97±20.47 pg/ml at 8h *p=0.01*, and 149.21±55.93 vs. 30.61±11.73 at 26h *p=0.01*). No differences on the P4 level between groups were found at any point. Exp.1 showed a significant difference in days to deliver after treatment: 2.84±1.02 in the E2 group vs 7.18±1.11 in the C group (p=0.01). Furthermore, the E2 treatment significantly impacted gene expression in all the tissues tested (p-value ≤ 0.1 and log2 fold change|0.5|). The cervix exhibited the strongest effect, with 8,073 DEGs, including E2 upregulation of *PTGS-2, PTGES2, ESR2*, and *IL-6*, and downregulation of *IL-10, ACTA2, MYH11*, *GJA1*, and *TIMPs*. The endometrium also showed significant changes, with 722 DEGs, including E2 upregulation of *OXTR* and *ESR1* and downregulation of *SULT1E1* and *TIMPs*. The caruncles had a more modest response, with 109 DEGs, while the myometrium was less responsive to the treatment, with 90 DEGs, including E2 upregulation of *PTGS-2*. Our findings suggest that E2 causes significant changes in gene expression in the maternal reproductive tissues, leading to parturition without a decline in circulating levels of P4. These effects appear to be primarily driven by the alterations in the cervix, where E2 upregulated genes associated with inflammation and cervical ripening and downregulated genes associated with smooth muscle contraction and tissue integrity.

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