**Materials and Methods SuppInfo**

*Sample preparation*

The myometrial sample was obtained from the superior aspect of the standard transverse incision. Tissues were washed with saline, snap frozen in liquid nitrogen, and stored at −80°C. An accurately weighed myometrial sample (30 mg) was transferred to a 1.5-mL Eppendorf tube. Two small steel balls were added to the tube. Then, 20 μL of internal standard (2-chloro-l-phenylalanine in methanol, 0.3 mg/mL) and 400 μL of extraction solvent with methanol/water (4/1, v/v) were added to each sample. Samples were stored at −80°C for 5 min, ground at 60 HZ for 2 min, subjected to ultrasonication in an ice water bath for 10 min, and then stored at −20°C for 30 min. The extract was centrifuged at 15000 rpm at 4°C for 10 min. The supernatants (200 μL) from each tube were collected using crystal syringes, filtered through 0.22 μm microfilters, and transferred to LC vials. The vials were stored at −80°C until LC-MS analysis. Quality control (QC) samples were prepared by pooling the aliquots of all the samples.

All chemicals and solvents were analytical or high-performance liquid chromatography (HPLC) grade. Water, methanol, acetonitrile, and formic acid were purchased from CNW Technologies GmbH (Düsseldorf, Germany). L-2-chlorophenylalanine was purchased from Shanghai Hengchuang Bio-technology Co., Ltd. (Shanghai, China).

*Mass spectrometry analysis*

A total of 60 myometrial samples were for metabolite profiling by a An ACQUITY UHPLC system coupled with a Xevo G2-XS mass spectrometer (LC-MS) approach (Waters Corporation, Milford, CT, USA). An ACQUITY UPLS BEH C18 column (1.7 μm, 2.1 × 100 mm) was employed in both positive and negative modes. The binary gradient elution system consisted of (A) water (containing 0.1% formic acid, v/v) and (B) acetonitrile (containing 0.1% formic acid, v/v). The linear gradient was as follows: 0 min, 1% B; 1 min, 30% B; 2.5 min, 60% B; 6.5 min, 90% B; 8.5 min, 100% B; 10.7 min, 100% B; 10.8 min, 1% B; and 13 min, 1% B. The flow rate was 0.4 mL/min, and the column temperature was 45°C. All the samples were kept at 4°C during the analysis. The injection volume was 2 μL.

The QCs were injected at regular intervals (every 10 samples) throughout the analytical run to provide a set of data from which repeatability could be assessed (Fig.1 SuppInfo).

*Data preprocessing and statistical analysis*

The acquired LC-MS raw data were analyzed by the progqenesis QI software (Nonlinear Dynamics, Newcastle, UK) using the following parameters. Precursor tolerance was set 5 ppm, fragment tolerance was set 10 ppm, and retention time (RT) tolerance was set 0.02 min. Internal standard detection parameters were deselected for peak RT alignment, isotopic peaks were excluded for analysis, and noise elimination level was set at 10.00, minimum intensity was set to 15 % of base peak intensity. The Excel file was obtained with three-dimension data sets including m/z, peak RT and peak intensities, and RT–m/z pairs were used as the identifier for each ion. The resulting matrix was further reduced by removing any peaks with missing value (ion intensity = 0) in more than 60 % samples. Qualitative identification of the compounds was conducted using The Human Metabolome Database, Lipidmaps (v2.3), and the METLIN database.

Next, the positive and negative data were combined to get a combine data which was imported into SIMCA software package (version 14.0, Umetrics, Umeå, Sweden). Principle component analysis (PCA) and (orthogonal) partial least-squares-discriminant analysis (O)PLS-DA were carried out to visualize the metabolic alterations among two groups, after mean centering (Ctr) and Pareto variance (Par) scaling, respectively. The Hotelling’s T2 region, shown as an ellipse in score plots of the models, defines the 95% confidence interval of the modeled variation. Variable importance in the projection (VIP) ranks the overall contribution of each variable to the OPLS-DA model, and those variables with VIP > 1 are considered relevant for group discrimination. In this study, the default 7-round cross-validation was applied with 1/seventh of the samples being excluded from the mathematical model in each round, in order to guard against overfitting. The differential metabolites were selected on the basis of the combination of a statistically significant threshold of variable influence on projection (VIP) values obtained from the OPLS-DA model and p values from a two-tailed Student’s t test on the normalized peak areas, where metabolites with VIP values larger than 1.0 and p values less than 0.05 were considered as differential metabolites.

The statistical significance of intergroup differences was evaluated using Student’s t test or Kruskal–Wallis test, when appropriate, for pairwise comparisons and because of non-normal distribution of variables. All statistical tests were two-sided, and the level of significance was recognized as P < 0.05. Data were analyzed using the IBM SPSS 22.0 software (SPSS Inc, Chicago, IL, USA). The trends between the duration of labor and specific steroid metabolites were fitted by the “LOESS” function of the R software.