Urinary Estrogen and Progesterone Metabolite Patterns in Ovulatory and Anovulatory Women

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Objective

To determine if monthly urinary patterns of estrogens and progesterone metabolites, collected as dried urine samples and measured with a validated GC-MS/MS assay, differ between women with laboratory evidence of ovulation and women with no laboratory evidence of ovulation.

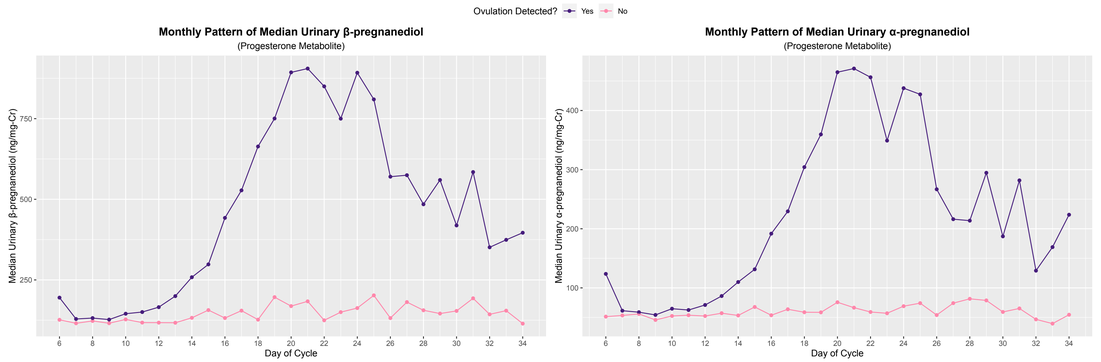
Materials and Methods

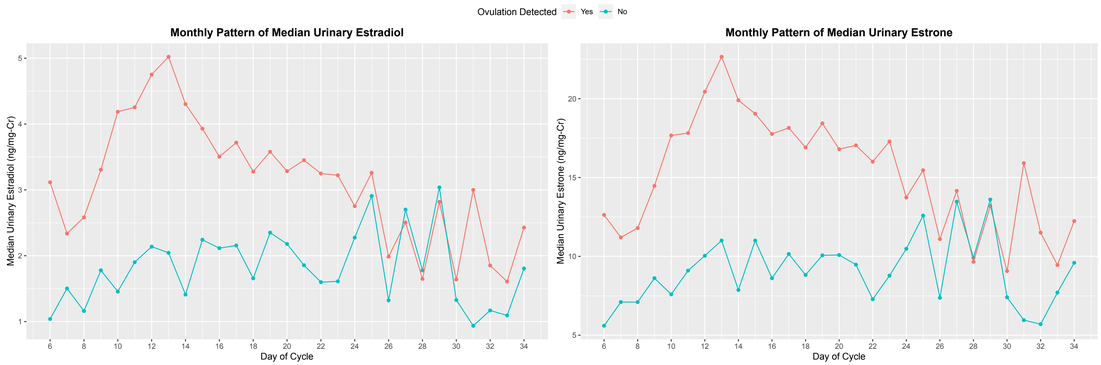
This was a retrospective observational cohort study conducted using a database containing 144,561 laboratory accessions that were submitted between January 1, 2016 and December 9, 2019 by 129,883 patients. These patients collected urine samples on filter paper at home and sent these collections to the laboratory to be processed. From this database, 1604 individuals completed a cycle collection and met inclusion criteria for the study (female sex, age between 17 and 50 years, body mass index [BMI] between 16 and 55 kg/m², and urinary creatinine > 0.1ng/mL). Progesterone was measured as its urinary progesterone metabolites 5α-pregnane-3α, 20α-diol (α-pregnanediol) and 5β-pregnane-3α, 20α-diol (β-pregnanediol), with total pregnanediols calculated as α-pregnanediol plus β-pregnanediol. Estrogen was also measured via the urinary metabolites with total estrogens calculated as the sum of all 10 measured metabolites. Ovulation was defined as a peak β-pregnanediol > 600 ng/mg-Cr and a peak α-pregnanediol > 200 ng/mg-Cr or a change in total pregnanediols > 650 ng/mg-Cr. Mixed models to account for repeated measures were used to compare hormone patterns between women who showed evidence of ovulation and those who did not.

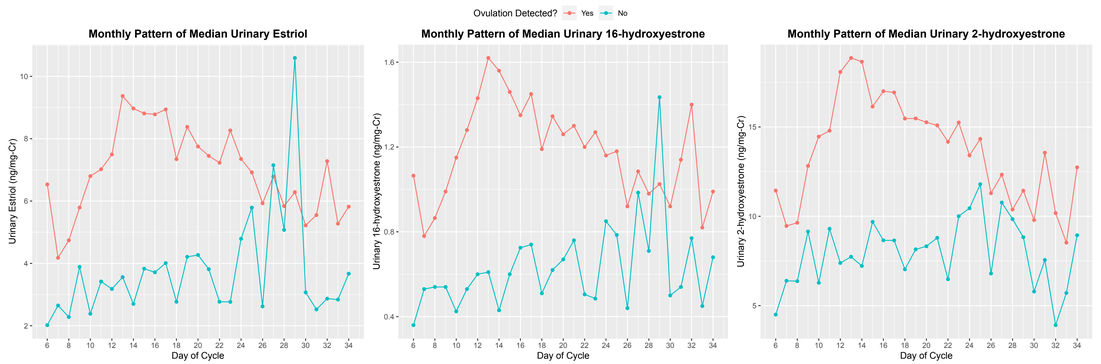
Results

Of the 1604 patients included in the study, 83% (1336) showed evidence of ovulation. The mean age (± SD) was 36.5 ± 6.8 for the group that showed evidence of ovulation and 34.3 ± 9.3 for the group that did not. The mean BMI was 24.1 ± 4.6 for the ovulation group and 24.6 ± 5.5 for the anovulatory group. No statistically significant difference existed between either the mean age (p = 0.15) or BMI (p = 0.43). A mixed model showed that the difference in the trajectories of total pregnanediols between those who ovulated and those who did not differed significantly (mean difference = 545.67 ± 28.2 ng/mg-Cr/day; p<0.0001). Similarly, in the mixed model evaluating differences in the patterns of total estrogens over the month, the trajectories differed between those who did and did not ovulate (mean Δ = 13.2±3.5 ng/mg-Cr/d). The individual pregnanediol and estrogen measures resulted in similar findings when analyzed separately.

Progesterone & Estrogen Metabolite Patterns







Subject Characteristics

Variable	Total	Ovulatory	Anovulatory	p value
Age (years)	36.1 ± 7.3	36.5 ± 6.8	34.3 ± 9.3	0.15
BMI (kg/m²)	24.2 ± 4.7	24.1 ± 4.6	24.6 ± 5.5	0.43
Peak α-pregnanediol (ng/mg-Cr)	560.4 (318.1, 882.8)	649.4 (441.2, 966.3)	98.8 (57.4, 173.0)	<0.0001
Peak β-pregnanediol (ng/mg-Cr)	1102.2 (678.0, 1606.0)	1233.9 (900.5, 1728.4)	256.6 (159.9, 384.0)	<0.0001

*Mean ± standard deviation for normally distributed variables; Median (IQR) for variables with a skewed distribution.

Conclusions

The method used in this study effectively captured the expected estrogen and progesterone metabolite patterns in women who showed laboratory evidence of ovulation. The results also showed clear and significant differences in these patterns between women who ovulated and women who did not. Further research comparing this method with more definitive methods of ovulation confirmation, such as ultrasonography, is needed.

Impact Statement

The results of this study demonstrate the potential for this tool to provide an easy to collect, lower cost, non-invasive option for clinicians and researchers investigating clinical scenarios involving ovulation status.