

EVALUATION OF BIOCHEMICAL HYPERANDROGENISM IN ADOLESCENT GIRLS WITH MENSTRUAL IRREGULARITIES

EVALUACIJA BIOHEMIJSKOG HIPERANDROGENIZMA KOD ADOLESCENTKINJA SA NEREDOVNIM MENSTRUALNIM CIKLUSOM

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Summary

Background: The aim of this study was to evaluate fertility hormone levels in adolescent girls and ten years older women with menstrual irregularities and with or without polycystic ovaries on ultrasound examination.

Methods: The study population consisted of 276 patients aged 12–18 years and 469 patients aged 22–28 years who had menstrual irregularities with or without polycystic ovaries on ultrasound examination. All subjects underwent a comprehensive medical assessment including documentation of the detailed history, physical and gynecological examination, measurement of the essential laboratory variables, and measurement of the fertility hormone levels.

Results: Within 745 patients (mean age: 21.4±4.8), 276 patients were aged 12–18 years (group 1) and 469 patients were aged 22–28 years (group 2). Dehydroepiandrosterone sulfate (DHEA-S) (237.7 (22.6–721.5) vs. 162.5 (2.4–660.7) respectively; $p < 0.001$) was significantly higher in group 1 than group 2. There were 74 subjects (26.8%) with LH/FSH ratio > 2 in group 1 and 74 subjects (15.8%) with LH/FSH ratio > 2 in group 2 ($p < 0.001$).

Conclusions: Biochemical hyperandrogenism is much more valuable in adolescents than in young adult women for the diagnosis of PCOS. Biochemical hyperandrogenism should be tested in adolescents with menstrual irregularities.

Keywords: polycystic ovary syndrome, DHEA-S, biochemical hyperandrogenism

Kratak sadržaj

Uvod: Cilj ove studije bio je da se odrede nivoi hormona koji utiču na fertilitet kod adolescentkinja i deset godina starijih žena sa neredovnim menstrualnim ciklusom sa ili bez ultrazvučnog nalaza o policističnim jajnicima.

Metode: Populacija u studiji sastojala se od 276 pacijentkinja uzrasta 12–18 godina i 469 pacijentkinja starosti 22–28 godina sa neredovnim menstrualnim ciklusom i sa ili bez ultrazvučnog nalaza o policističnim jajnicima. Sve ispitanice podvrgnute su sveobuhvatnoj medicinskoj proveri koja je uključivala dokumentaciju o detaljnoj istoriji, fizički i gineološki pregled, merenje osnovnih laboratorijskih varijabli i merenje nivoa hormona koji utiču na fertilitet.

Rezultati: Od 745 pacijentkinja (prosečna dob: 21,4±4,8), 276 pacijentkinja imalo je između 12 i 18 godina (grupa 1) a 469 pacijentkinja imalo je između 22 i 28 godina (grupa 2). Dehidroepiandrosteron sulfat (DHEA-S) (237,7 (22,6–721,5) vs. 162,5 (2,4–660,7); $p < 0,001$) bio je značajno viši u grupi 1 nego u grupi 2. U grupi 1, 74 ispitanice (26,8%) imale su odnos LH/FSH > 2 , dok su u grupi 2 takođe 74 ispitanice (15,8%) imale odnos LH/FSH > 2 ($p < 0,001$).

Zaključak: Biohemijski hiperandrogenizam ima mnogo veću vrednost kod adolescentkinja nego kod mladih žena za dijagnostikovanje sindroma policističnih jajnika. Biohemijski hiperandrogenizam treba testirati kod adolescentkinja sa neredovnim menstrualnim ciklusom.

Ključne reči: sindrom policističnih jajnika, DHEA-S, biohemijski hiperandrogenizam

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Introduction

Polycystic ovary syndrome (PCOS) is the most common endocrinological disease in reproductive age women with the prevalence between 6 and 15% depending on different diagnostic criteria (1). According to a consensus meeting held in Amsterdam in 2012, three of the Rotterdam criteria should be included for the diagnosis of PCOS in adolescents which are chronic anovulation, clinical or biochemical hyperandrogenism and polycystic ovary appearance. Polycystic ovary morphology (PCOM) is defined as >12 follicles with the diameter of 2–9 mm in ovaries or ovary volume of >10 mL on pelvic ultrasonography (2). Because multicystic ovaries are normal during pubertal development, PCOM is not accepted as a diagnostic criterion in adolescents. The Endocrine Society Clinical Practical Guideline (2013) suggested that PCOS can be diagnosed with the presence of clinical and/or biochemical evidence of hyperandrogenism and chronic anovulation in adolescents (3).

As PCOS is associated with significant reproductive morbidity as shown by the high prevalence of anovulatory infertility, spontaneous abortion, gestational diabetes and preeclampsia, early diagnosis in this case is very important (4). Among the women of reproductive age, these risks are strongly linked to insulin resistance and are compounded by the common occurrence of obesity (5). Although PCOS is reported to be a growing problem among the adolescent girls, not much data are available on the prevalence and characteristics of PCOS in developing countries, especially among the adolescent and young adults. Hence, PCOS is rarely diagnosed in the early teenage years and the hyperandrogenic effects usually progress slowly over time. It is not uncommon for girls with PCOS to have normal appearing ovaries but still have an imbalance in their hormone levels and, hence, the diagnosis of PCOS is often missed in this age group (6). Several fertility hormones such as follicle stimulating hormone (FSH), luteinizing hormone (LH), estradiol (E2), prolactin (PRL), total testosterone, thyroid stimulating hormone (TSH), free thyroxine (T4), dehydroepiandrosterone sulfate (DHEA-S), 17-OH progesterone and androstenedione can be tested for evaluating the patients with PCOS.

The aim of this study was to evaluate fertility hormone levels in adolescent girls and ten years older women with menstrual irregularities and with or without polycystic ovaries on ultrasound examination.

Materials and Methods

The study population consisted of 276 patients aged 12–18 years and 469 patients aged 22–28 years who consulted with menstrual irregularities between January 2013 and March 2016 with or without polycystic ovaries on ultrasound examination. The subjects had not previously been treated with fertility hormone therapy. All subjects had menarche for at least 2 years.

Eligible subjects underwent a comprehensive medical assessment including documentation of the detailed history, physical and gynecological examination, measurement of the essential laboratory variables, and measurement of the fertility hormone levels. Exclusion criteria were malignancy (all types of malignancy including basal cell carcinoma), presence of ovarian mass or cyst, and any other endocrinological disorders such as Cushing disease, Addison disease, thyroid disease, hyperprolactinemia, biochemical hyperandrogenism. Patients who were already receiving any hormone medicine were also excluded. All women in our unit routinely gave informed consent for the use of their data for research purposes. The possibility of receiving subsequent phone calls to ascertain follow up is also stated in this informed consent, which has been signed by all participating women.

Fasting blood samples were drawn from the antecubital vein from the participants after they had fasted > 12 hours on the third to fifth day of menstrual cycle. Patients avoided high stress levels and strenuous exercise because hormones, especially prolactin, are sensitive to these conditions. Blood samples were allowed to clot for 20–30 minutes at room temperature and then centrifuged at $3000 \times g$ for 5 minutes and frozen to below -80°C until analysis. Serum FSH, LH, Estradiol (E2), total testosterone, Prolactin, TSH, free thyroxine (T4), and DHEA-S were measured by an electrochemoluminescent immunoassay method (Konelab). LH/FSH ratio was calculated for each patient.

Statistical analysis

Data are demonstrated as mean \pm SD for normally distributed continuous variables, median (minimum–maximum) for skew distributed continuous variables, and frequencies for categorical variables. Pearson chi-square test was performed for the comparison of categorical variables. Means of normally distributed continuous variables were compared by ANOVA. Skew distributed continuous variables were compared by Mann Whitney U test. Pearson and Spearman analysis was used to identify correlations between study parameters. Statistical Package for Social Sciences (SPSS) for Windows version 22.0 (SPSS Inc., Chicago) was used for the analysis and two sided p value of < 0.05 was considered as significant.

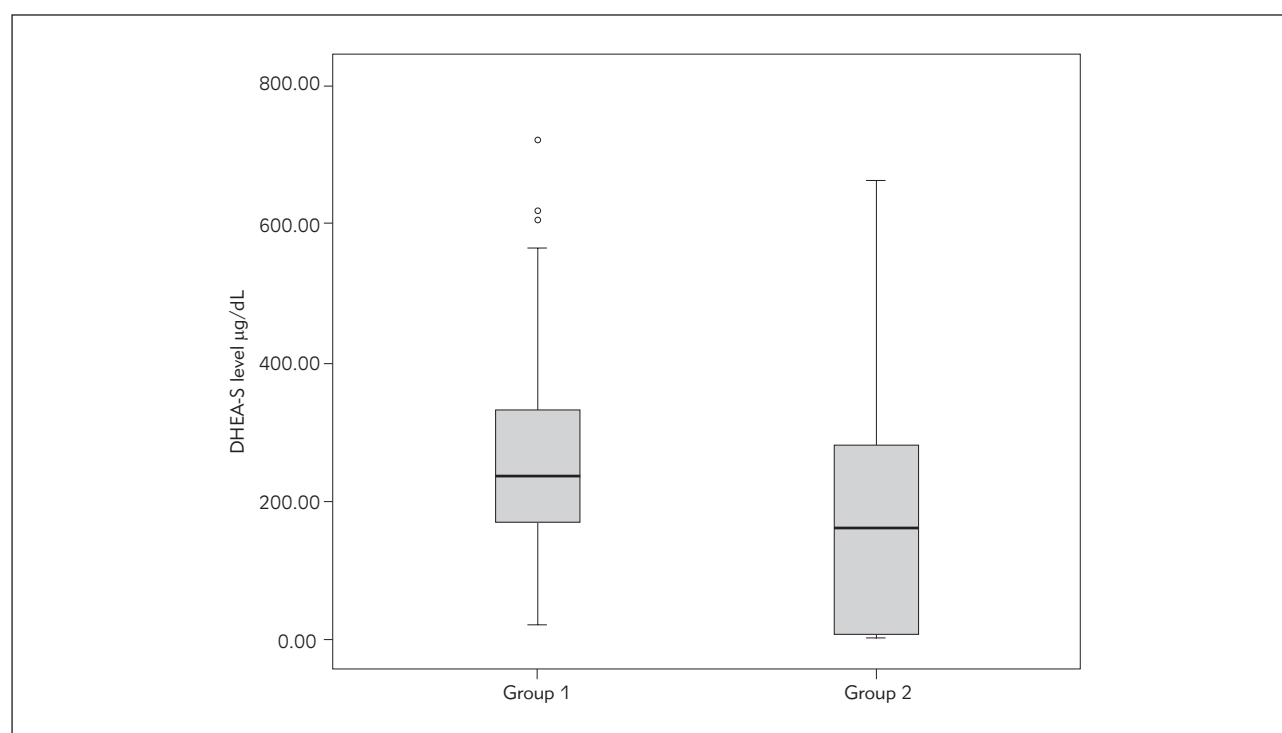
Results

Within 745 patients (mean age: 21.4 ± 4.8), 276 patients were aged 12–18 years (group 1) and 469 patients were aged 22–28 years (group 2). The mean age was 15.5 ± 1.4 in group 1, 24.9 ± 1.9 in group 2.

Dehydroepiandrosterone sulfate (DHEA-S) (237.7 (22.6 – 721.5) vs. 162.5 (2.4 – 660.7) $\mu\text{g/dL}$ respectively; $p < 0.001$) and Prolactin (15.5 (0.22 – 138.2) vs. 12.9 (1.41 – 149.0) ng/mL respectively;

Table I Comparison of patients according to hormone levels.

	12–18 aged group (no.276)	22–28 aged group (no.469)	<i>p</i>
FSH (mIU/mL)	5.6 (0.1–78.6)	6.2 (1.6–90.0)	0.001
LH (mIU/mL)	6.6 (0.01–72.09)	6.3 (0.51–95.63)	NS
E2 (pg/mL)	39 (3–579)	41 (5–507)	NS
TSH (mIU/mL)	1.9 (0.53–10.16)	1.8 (0.01–42.68)	NS
PRL (ng/mL)	15.5 (0.22–138.2)	12.9 (1.41–149.0)	0.001
Total testosterone (ng/mL)	0.35 (0–1.12)	0.35 (0.02–1.53)	NS
DHEA-S (mg/dL)	237.7 (22.6–721.5)	162.5 (2.4–660.7)	<0.001
LH/FSH ratio	1.18 (0.03–6.64)	1.02 (0.17–7.98)	NS

**Figure 1** Distribution of DHEA-S levels in group 1 and 2.

$p=0.001$) levels were significantly higher in group 1 than group 2. Follicle stimulating hormone (FSH) (5.6 (0.1–78.6) vs. 6.2 (1.6–90.0) mIU/mL respectively; $p=0.001$) level was significantly lower in group 1 than group 2. There was no significant difference in luteinizing hormone (LH) (6.6 (0.01–72.09) vs. 6.3 (0.51–95.63) mIU/mL respectively $p=NS$), estradiol (E2) (39 (3–579) vs. 41 (5–507) pg/mL respectively $p=NS$), LH/FSH ratio (1.18 (0.03–6.64) vs. 1.02 (0.17–7.98) respectively; $p=0.058$), total testosterone (0.35 (0–1.12) vs. 0.35 (0.02–1.53) ng/mL respectively $p=NS$), thyroid stimulating hormone (TSH) (1.9 (0.53–

10.16) vs. 1.8 (0.01–42.68) μ IU/mL respectively $p=NS$) levels between two groups (Table I).

There were 74 subjects (26.8%) with LH/FSH ratio >2 in group 1 and 74 subjects (15.8%) with LH/FSH ratio >2 in group 2 ($p<0.001$). Bivariate correlation analysis showed that DHEA-S was positively correlated with total testosterone ($r=0.450$, $p<0.001$) and PRL ($r=0.088$, $p=0.016$) levels and negatively correlated with age ($r=-0.247$, $p<0.001$). There was no correlation between DHEA-S level and LH/FSH ratio and any other clinical and laboratory parameters.

Discussion

This study evaluated biochemical hyperandrogenism in adolescent girls and young adult women who have menstrual irregularities and with or without polycystic ovaries on ultrasound examination. Despite the ongoing debate on the diagnosis of PCOS in adolescent girls, current guidelines emphasize the importance of elevated androgens.

In this study, we demonstrated that DHEA-S levels were higher in adolescent girls than young adult women. The study also showed that prolactin levels were higher in adolescent girls than young adult women.

The main clinical presentation in adolescents with PCOS is menstrual irregularity which is mostly oligomenorrhea. It is important to distinguish these irregularities observed in the first 2–3 years after menarche because of the immaturity of the hypothalamo-hypophyseal-ovarian axis (7). Adolescent girls in this study were selected from subjects having menarche for at least 2 years.

Another difficulty for the diagnosis of PCOS in adolescents is the ultrasonographic appearance of ovaries. Ovarian volume and follicle number and size are assessed to define PCOS with ultrasonography. Although polycystic ovary morphology and increased ovarian volume could be diagnostic for PCOS in adult women, this appearance is observed normally in adolescent girls. Furthermore, there is another limitation about the utility of ultrasonography in this age group. There is the necessity of transvaginal ultrasonography especially in obese girls who are a large portion of adolescents with PCOS. Therefore, ultrasonography is not a first step investigation in adolescents. In our study, adolescents with menstrual irregularities had high androgen levels independent from the presence of polycystic ovary morphology.

Hyperandrogenism is clinically manifested as acne, hirsutism or alopecia which are commonly ob-

served during normal pubertal period. These clinical findings are not helpful for the diagnosis of PCOS in adolescent girls. Furthermore, Ferriman Gallwey scoring system cannot be applied to adolescents who have not completed pubertal development. Hence, biochemical hyperandrogenism has been proposed to be the most important finding for the diagnosis of PCOS in this age group (8). According to the international pediatric subspecialty societies, unexplained persistent anovulation and biochemical hyperandrogenism can be used as diagnostic criteria for PCOS in adolescents (9). Patients with PCOS have elevated androgen levels throughout life (10). Comparing to women with normal menses, patients with PCOS have increased serum LH levels, low-normal FSH levels and increased LH/FSH ratios depending on the feedback mechanisms. In PCOS, LH/FSH ratio is generally higher than 2 (11, 12). In our study, there were much more subjects who had LH/FSH ratio >2 in adolescents than young adult woman.

Although there is still ongoing debate which diagnostic criteria should be used, PCOS is a diagnosis of exclusion. It is important to avoid the overdiagnosing or underdiagnosing of PCOS with exclusion of other endocrinological or tumoral conditions. Early diagnosis and management of PCOS in adolescents should be necessary due to association with metabolic outcomes in the future (12, 13).

In conclusion, biochemical hyperandrogenism is much more valuable in adolescents than in young adult women for the diagnosis of PCOS. Biochemical hyperandrogenism should be tested in adolescents with menstrual irregularities.

Conflict of interest statement

The authors stated that they have no conflicts of interest regarding the publication of this article.

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Received: April 14, 2017

Accepted: June 22, 2017