Available online at **www.ijmrhs.com**

ISSN No: 2319-5886

International Journal of Medical Research & Health Sciences, 2018, 7(7): 36-41

Menstrual Disorders and Endometrial Thickness in Adolescents: Endocrinological and Sonographic Approach

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ABSTRACT

Objective: *The research is aimed at identifying those risk factors in adolescence which is responsible for diseases in adulthood, and to delineate the endocrinological profile in adolescents with persistent irregular menstrual periods in association with pelvic ultrasound examination.* ***Methods:*** *Our study population included girls who were admitted to the endocrinology and gynecology unit in the Department of Pediatric, L'Aquila, Italy, from March 2016 to January 2017. Our study population included 80 girls, 40 had oligomenorrhea (Group "Olig") and 40 had no menstrual disorders (Group "Eumen"). At the basal time, we obtained a complete dosage of hormones and investigated metabolic profile. Between the 5th and the 10t^{h} day of the spontaneous menstrual cycle we performed a pelvic ultrasound and on the 28th day for the group with "Olig" and on the 14th day for the "Eumen" group, we dosed serum LH and 17-estradiol. Also on the 21st day and on the 35th day, we obtained a dosage of serum progesterone.* ***Results:*** *Pituitary gonadotropins dosages revealed in the periovulatory phase low LH values in the group "Olig", also associated with lower estradiol values. In addition, luteal phase progesterone showed adequate values only in the "Eumen" group. Moreover, the execution of pelvic ultrasound revealed a statistically significant difference in the endometrial thickness to be associated with estrogenic action not contrasted with progesterone.* ***Conclusions:*** *In our study, we found low LH values in girls with oligomenorrhea with lower estradiol values, which was unable to create positive feedback for the peak of LH. Pelvic ultrasound has highlighted an increased endometrial thickness in presence of persistent menstrual disorders.*

Keywords: Adolescents, Oligo, Anovulation, Endometrial thickness, Endometrial hyperplasia, Pelvic ultrasound

INTRODUCTION

Menstrual disorders in adolescents are a common problem in clinical practice, representing an important source of anxiety for patients and their parents [1]. Around 80% of adolescents experience irregular menstrual periods during the first 2 years after menarche because the transition process of hypothalamic-pituitary-ovarian (HPO) axis into adult typical pattern generally acts gradually [2,3]. For these reasons, in this age, menstrual periods tend to have their own characteristics, which can differ widely from those of adult women [4]. Furthermore, in adolescents, anovulation is frequently due to a persistent elevation, failure to reach adequate estrogen levels, due to immature HPO axis, which can inhibit ovulation through the disruption of mechanisms proposed to luteinizing hormone (LH) surge [5,6]. Apart from such physiological aspect, menstrual intervals persistently greater than 45 days during 2 or more years after menarche, represent signs of oligo-anovulation [7]. When anovulation occurs, unopposed estrogen stimulation of endometrium may result in endometrial and glands proliferation, with pseudostratification and mitotic activity [8]. Uterine cancer is the 7th leading cause of death from cancer among women [9] and, approximately 80% of endometrial cancers are likely due to hormonal imbalances [10]. The 14% of women with recurrent anovulatory cycles develop cancer precursors (hyperplasia with atypia) and 5-10% of these progresses to endometrial carcinoma [11-13]. Endometrial cancer is rare in adolescents but should be taken into consideration as a potential risk factor in cases with persistent anovulation and young girls with individual risk factors [9]. Although polycystic ovary syndrome (PCOS) is the most common cause of anovulation, nevertheless it is not the only one, and once the diagnosis of PCOS has been excluded, according to

androgen excess and PCOS society criteria of 2006, menstrual alterations should be strictly investigated, particularly in cases of precocious menarche and who are having a family history of endometrial hyperplasia or cancer [14].

Pelvic Ultrasound in Adolescents

Pelvic ultrasound with the abdominal approach is a very important weapon to analyze gonads and uterus. Its clinical relevance extends to all phases of women's reproductive life [15]. In adolescents with irregular periods, the ultrasound study of uterus and ovaries morphology, with cyclic changes of the endometrium, would be helpful [16]. Several studies have highlighted the association between unopposed estrogen stimulation and an increase of endometrial thickness, particularly in women with PCOS [17]. Normal endometrial thickness depends on the menstrual cycle phase: a thickness greater than 15 mm is considered normal in the secretory phase, while hyperplasia, assessed during the early proliferative phase, can be reliably excluded only if the endometrium thickness is less than 7-9 mm, as suggested by the retrospective study [18]. The increased thickness derives from progestogen deficit as a result of anovulation. In this hormonal environment, endometrium can further develop, without presenting the typical morphology periovulatory and postovulatory. For a correct evaluation of endometrium, ultrasound study should be performed at day 5^{th}-10^{th} of the menstrual cycle, to reduce the wide physiological variation in endometrial thickness and appearance, observed in the proliferative phase [19]. In our study, since there are no validated data on endometrial cut-off in adolescence, we compared our ultrasound data with a control group, including adolescents without menstrual irregularities, in order to identify the differences in endometrial thickness and eventually select cases that may require a different approach.

PATIENTS AND METHODS

Our study population included 80 girls who were admitted to the endocrinology and gynecology unit in the Department of Pediatric, L'Aquila, Italy, from March 2016 to January 2017. Of these, 40 had oligomenorrhea (Group "Olig") and 40 had no menstrual disorders (Group "Eumen").

Inclusion Criteria

Adolescent's girls aged between 14 and 18 years, the persistent presence of oligomenorrhea from the menarche, menarchal age over four years, and body mass index within the normal range by gender and age were included in the study.

Exclusion Criteria

Girls with confirmed PCOS and girls with hirsutism or biochemical hyperandrogenemia in a previous assessment, cancer, chronic illnesses, or other endocrinological disorders (e.g. epilepsy, chronic inflammatory disease), suspected or confirmed eating disorders, according to diagnostic and statistical manual of mental disorders (DSM-5) and to appropriate testing (SCOFF), were eventually excluded from the study [20].

Data and Sample Collection

Significant familiar history (PCOS, family history of endometrial or breast cancer), personal history (age of menarche, another previous endocrinological disease, rapid change in body weight), clinical and previous laboratory data were collected in a medical record for each patient of both groups. During the first visit, a global physical examination, calculation of body mass index percentile (according to SIEDP charts) and Ferriman-Gallwey scale were performed. Further information was obtained on menstrual cycles (about the first cycle and periodicity, frequency, length, flow, dysmenorrhea, associated symptoms in the subsequent cycles). Furthermore, during inter-menstrual period, blood tests for hematological and coagulation status, free thyroxine (fT4), thyroid stimulating hormone (TSH), prolactin (PRL), dehydroepiandrosterone sulphate (DHEAS), delta 4-androstenedione (Δ4A), total testosterone (TT), free testosterone (FT) and an oral glucose tolerance test (OGTT) were performed in order to rule out other endocrinological conditions which could cause menstrual disorders. Subsequently, each girl, after the reappearance of the menstrual cycle, went to the medical clinic, to undergo the medical scheme, as follows:

- Between the 5^{th} and the10^{th} day to perform a pelvic ultrasound during the follicular phase.
- On the 28^{th} day for the group with oligomenorrhea and on the 14^{th} day for the control group to obtain LH and 17-estradiol blood levels.
- On the 21^{st} day and on the 35^{th} day, respectively, to obtain progesterone blood level.

Laboratory Measurements

The blood sampling, obtained by venipuncture with aseptic technique, was performed in fasted patients for at least 8 hours. To investigate hormonal profile, LHRH test was administered, injecting an IV bolus of 100 μgr/m^2 of Luteinizing Hormone Releasing Factor, (Lutrelef®) to simultaneously dose LH and FSH hormones at time 0, 10', 20', 40' and 60' after the stimulus. For the determination of LH, ARCHITECT LH® test, a two-step chemiluminescent microparticle immunoassay (CMIA) for the quantitative determination of human luteinizing hormone (LH) in human serum and plasma, was used. The test automatically calculates the hormone's concentrations (expressed in mIU/mL), measuring between 0.09 and 250 mIU/mL, based on the inter-assay precision of a CV (coefficient of variation) of 22% (functional sensitivity). For the determination of FSH, the ARCHITECT FSH® test, a two-step chemiluminescent microparticle immunoassay (CMIA), was used. The test automatically calculates the FSH's concentrations (expressed in mIU/mL), measuring between 0.00 and 150 mIU/mL, based on the inter-assay precision of a CV (coefficient of variation) of 10% (functional sensitivity). For the determination of DHEA-S, the ARCHITECT DHEA-S® test, a delayed one step chemiluminescent microparticle immunoassay (CMIA), was used. The test automatically calculates the hormone's concentrations (expressed in μg/dL), measuring between 3.0 and 1500 μg/dL, based on the inter-assay precision of a CV (coefficient of variation) of 10% (functional sensitivity). For the determination of testosterone, the ARCHITECT 2nd generation testosterone® test, a delayed one-step chemiluminescent microparticle immunoassay (CMIA), was used. The test automatically calculates the testosterone's concentration (expressed in nmol/L), measuring between 0.03 and 35 nmol/L, based on the inter-assay precision of a CV (coefficient of variation) of 10% (functional sensitivity). For the determination of Estradiol, the ARCHITECT Estradiol® test, a delayed one step chemiluminescent microparticle immunoassay (CMIA), was used. The test automatically calculates the hormone's concentrations (expressed in nmol/L), measuring between 0.00 and 70.0 nmol/L, based on the inter-assay precision of a CV (coefficient of variation) of 20% (functional sensitivity). For the determination of DHEA-S, the ARCHITECT DHEA-S® test, a delayed one-step chemiluminescent microparticle immunoassay (CMIA), was used. The test automatically calculates the hormone's concentrations (expressed in μg/dL), measuring between 3.0 and 1500 μg/dL, based on the inter-assay precision of a CV (coefficient of variation) of 10% (functional sensitivity). The determination of Delta-4-androstenedione was obtained by the IMMULITE 2000® test, a chemiluminescent microparticle immunoassay (CMIA). The test automatically calculates the hormone's concentration (expressed in ng/mL), measuring between 0.3 and 10 ng/mL, based on the inter-assay precision of a CV (coefficient of variation) of 10% (functional sensitivity). To investigate the metabolic profile, an OGTT (oral glucose tolerance test) was performed (1.75 g/kg, max 75 g pro capite) and plasma glucose and insulin obtained were utilized to calculate the glucose/insulin ratio (glucose/insulin <6: insulin resistance). The insulin resistance was estimated using the homeostasis model assessment (HOMA-IR); values>2.5 indicating insulin resistance.

Ultrasound Examination

Ultrasound was performed with an abdominal approach using a LOGIQ P9, GE Healthcare® using a 3.5 MHz curvilinear probe, obtaining a complete scan through the uterus and adnexal region in both the longitudinal and transverse planes. Uterine and ovarian volumes were calculated with the appropriate program automatically. Endometrium measurement corresponded to the distance between the two basal layers of the anterior and posterior uterine walls at the echogenic interface between endometrium and myometrium was performed.

Consent

Written informed consent of the patient's data was obtained from the parents or guardian of the patients. Their data are anonymous.

Non-Parametric Statistical Analysis

All statistical analysis was performed using R-statistical software (version 2.14.0; R Foundation for Statistical Computing, Vienna, Austria) [21]. Data were expressed as the mean ± standard deviation (SDS). Statistical analysis was performed using one sample student t-test. A value of $p<0.05$ was considered statistically significant.

RESULTS

Patients of the 2 study groups were homogeneous for age, menarche, menarchal age and BMI at observation time (Table 1).

Table 1 Population age and body mass index

Variables	Group Olig (n:40)	Group Eumen (n:40)	Statistical Analysis
Age years (SDS)	16.5 (1.2)	16.2 (1.28)	NS
BMI kg/m^2 (SDS)	19.7 (2.17)	20.25 (2.04)	NS
NS: Not significant			

Data collected over the menstrual cycle showed a significant difference in the length of the menstrual cycle (45.7 days vs. 30.5 days) with a bleeding duration overlapping between the two groups (5.75 vs. 5.65 days) (Table 2).

Table 2 Cycle characteristics

Variables	Group Olig (n:40)	Group Eumen (n:40)	Statistical Analysis
Age of menarchal years (SDS)	11.7 (1.14)	12.0 (1.05)	NS
Menarchal age years (SDS)	4.45 (0.75)	4.35 (0.58)	NS
Cycle interval days (SDS)	45.75 (9.14)	30.55 (4.05)	t =-6.7928, df=26.201, p-value=3.159, e-07, p<0.05
Flow length days (SDS)	5.75 (1.25)	5.65 (1.18)	NS
NS: Not significant			

Thyroid profiles, prolactin, total and fractional testosterone, DHEAS, androstenedione, 17OHP, and HOMA-IR were found to be in the standard range in both groups. Pituitary gonadotropins dosages revealed statistically significant differences between FSH and LH, detected in the late follicular/periovulatory phase with low LH values in the group with oligomenorrhea, also associated with lower estradiol values with respect to the group with regular menstrual cycles. In addition, luteal phase progesterone showed adequate values (>3 ng/ml) only in the "Eumen" group being low in the "Olig" group, consistent with the low LH and E2 values (Table 3).

Table 3 Hormonal examination

Variables	Group Olig (n:40)	Group Eumen (n:40)	Statistical Analysis
TSH microUI/mL (SDS)	1.796 (0.751)	1.746 (0.754)	NS
FT4 ng/dl (SDS)	2.460 (0.803)	2.434 (0.819)	NS
PRL ng/ml (SDS)	11.940 (4.36)	12.834 (4.93)	NS
FSH mIU/ml (SDS)	5.348 (0.82)	3.89 (0.90)	t =-5.3208, df=37.628, p-value=4.986e-06, p<0.05
Total testosterone ng/dl (SDS)	21.90 (10.8)	24.8 (12.1)	NS
Free testosterone ng/dl (SDS)	1.928 (1.4)	1.814 (1.33)	NS
DHEAS mcg/dl (SDS)	171.3 (42.2)	156.6 (41.4)	NS
Androstenedione ng/dl (SDS)	177.5 (46.4)	174.4 (40.1)	NS
17OHP ng/dl (SDS)	1.043 (0.3)	1.052 (0.28)	NS
LH* mIU/ml (SDS)	2.072 (0.52)	7.546 (1.5)	t=-14.628, df=23.127, p-value=3.522e-13, p<0.05
17beta-estradiol* pg/ml (SDS)	60.6 (6.8)	108.0 (34.6)	t=5.9659, df=20.9, p-value=6.512e-06, p<0.05
Progesterone** ng/ml (SDS)	1.245 (0.34)	3.354 (1.54)	t=5.9659, df=20.9, p-value=6.512e-06, p<0.05
NS: Not significant; *Examination in the 28th day for the group with oligomenorrhea and the 14th day for the group with eumenorrhea; **Examination in the 21st day for the group with eumenorrhea oligomenorrhea and the 35th day for a group			

Moreover, the execution of pelvic ultrasound revealed a statistically significant difference in endometrial thickness, which is reported in the literature, to be associated with estrogenic action not contrasted with progesterone. The remaining ultrasound findings were normal in both groups (Table 4, Figure 1). The limits of our study are represented by the shortage of the sample, the lack of continuous LH monitoring, and the lack of multiple observations.

Table 4 Ultrasound pelvic values between the 5-10th day of the menstrual cycle

Variables	Group Olig (n:40)	Group Eumen (n:40)	Statistical Analysis
Uterine volume ml (SDS)	55.6 (6.2)	51.45 (11.8)	NS
Ovarian volume ml (SDS)	6.3 (1.05)	9.442(1.4)	NS
Endometrial thickness mm (SDS)	9.325 (1.086)	5.470 (1.14)	t=-10.913, df=37.891, p-value=2.959e-13, p<0.05
NS: not significant			

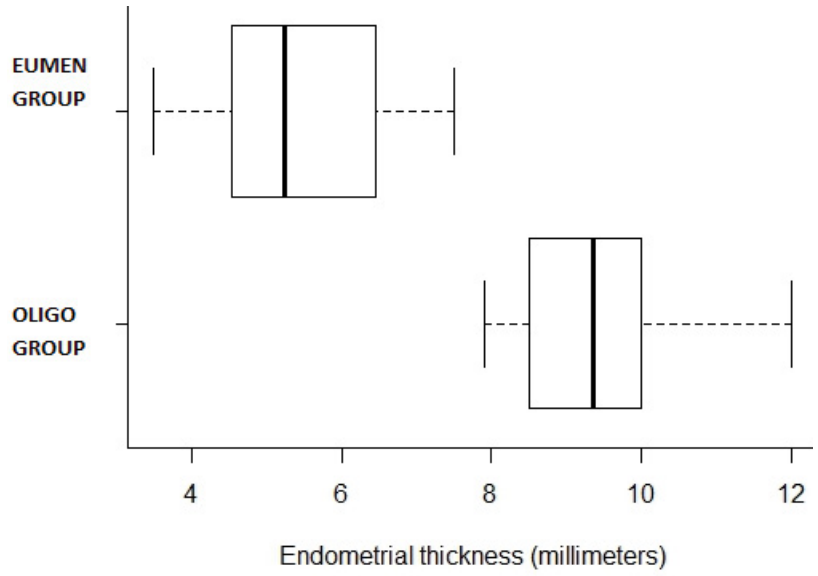

Figure 1 Endometrial thickness in the two groups

DISCUSSION

In our study, we found low LH values in girls with oligomenorrhea with lower estradiol values, unable to create positive feedback for the peak of LH, responsible for ovulation. FSH values were higher in girls with anovulation for lack of negative feedback.

In addition, progesterone values confirmed the lack of ovulation in the group with oligomenorrhea. In adolescents with a menarchal age greater than 2 years, menstrual cycle rhythm disorders should not be considered physiologic and should always be investigated.

Chronic anovulation is one of the risk factors for endometrial disease and reduced fertility, for this reason, all young women who complain of menstrual cycle disorders should be identified and monitored.

In particular, the use of pelvic ultrasound imaging with a simple transabdominal approach has highlighted an increased endometrial thickness compared to the control group, worthy of follow-up over time and of any pharmacological and/or biopsy approach if persistent. For these reasons, it is necessary to establish an etiological diagnosis of menstrual disorders in adolescence and to evaluate its persistence beyond the early menarchal age.

CONCLUSION

A low LH value in girls with oligomenorrhea with lower estradiol values were found, which was unable to create positive feedback for the peak of LH. Pelvic ultrasound has highlighted an increased endometrial thickness in presence of persistent menstrual disorders. Prevent by monitoring the risk factors in adolescents serious pathologies that arise in adults as in this case endometrial cancer.

DECLARATIONS

Conflict of Interest

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

REFERENCES

[1] World, Health Organization Task Force on Adolescent Reproductive Health. "World Health Organization multicenter study on menstrual and ovulatory patterns in adolescent girls." *Journal of Adolescent Health,* Vol. 7, No. 4, 1986, pp. 229-35.

[2] March, Charles M., et al. "Roles of estradiol and progesterone in eliciting the midcycle luteinizing hormone and

follicle-stimulating hormone surges." *The Journal of Clinical Endocrinology and Metabolism,* Vol. 49, No. 4, 1979, pp. 507-513.

[3] American Academy of Pediatrics Committee on Adolescence. "Menstruation in girls and adolescents: using the menstrual cycle as a vital sign." *Pediatrics,* Vol. 118, No. 5, 2006, p. 2245.

[4] Fraser, Ian S., et al. "The FIGO recommendations on terminologies and definitions for normal and abnormal uterine bleeding." *Seminars in Reproductive Medicine,* Vol. 29, No. 5, 2011.

[5] Direito, Ana, et al. "Relationships between the luteinizing hormone surge and other characteristics of the menstrual cycle in normally ovulating women." *Fertility and Sterility,* Vol. 99, No. 1, 2013, pp. 279-85.

[6] Zhang, Katie, et al. "Onset of ovulation after menarche in girls: a longitudinal study." *The Journal of Clinical Endocrinology and Metabolism,* Vol. 93, No. 4, 2008, pp. 1186-94.

[7] Munro, Malcolm G., et al. "FIGO classification system PALM□COEIN for causes of abnormal uterine bleeding in nongravid women of reproductive age." *International Journal of Gynecology and Obstetrics,* Vol. 113, No. 1, 2011, pp. 3-13.

[8] Maksem, John A., et al. *Endometrial cytology with tissue correlations*. Vol. 7. Springer Science and Business Media, 2009.

[9] Miller, Kimberly D., et al. "Cancer treatment and survivorship statistics, 2016." *CA: A Cancer Journal for Clinicians,* Vol. 66, No. 4, 2016, pp. 271-89.

[10] Carlson, Matthew J. "Catch it before it kills: progesterone, obesity, and the prevention of endometrial cancer." *Discovery Medicine* 14.76 2012: 215.

[11] Ash, Stephanie J., Scott A. Farrell, and G. Flowerdew. "Endometrial biopsy in DUB." *The Journal of Reproductive Medicine* 41.12 1996: 892-896.

[12] Soliman, Pamela T., et al. "Risk factors for young premenopausal women with endometrial cancer." *Obstetrics and Gynecology,* Vol. 105, No. 3, 2005, pp. 575-80.

[13] Lacey Jr, J. V., et al. "Endometrial carcinoma risk among women diagnosed with endometrial hyperplasia: the 34-year experience in a large health plan." *British Journal of Cancer,* Vol. 98, No. 1, 2008, p. 45.

[14] Fiederling, Jonas, Ahmad Zia Shams, and Ulrike Haug. "Validity of self□reported family history of cancer: A systematic literature review on selected cancers." *International Journal of Cancer,* Vol. 139, No. 7, 2016, pp. 1449-60.

[15] Persadie, Richard J. "Ultrasonographic assessment of endometrial thickness: a review." *Journal of Obstetrics and Gynaecology Canada,* Vol. 24, No. 2, 2002, pp. 131-36.

[16] Fleischer, Arthur C. "Sonographic assessment of endometrial disorders." *Seminars in Ultrasound, CT and MRI*, Vol. 20. No. 4. Elsevier, 1999.

[17] Cheung, Anthony P. "Ultrasound and menstrual history in predicting endometrial hyperplasia in polycystic ovary syndrome." *Obstetrics and Gynecology,* Vol. 98, No. 2, 2001, pp. 325-31.

[18] McCormick, Betsy A., et al. "Endometrial thickness predicts endometrial hyperplasia in patients with polycystic ovary syndrome." *Fertility and Sterility,* Vol. 95, No. 8, 2011, pp. 2625-27.

[19] Nalaboff, Kenneth M., John S. Pellerito, and Eran Ben-Levi. "Imaging the endometrium: disease and normal variants." *Radiographics,* Vol. 21, No. 6, 2001, pp. 1409-24.

[20] Sanchez-Armass, Omar, et al. "Validation of the SCOFF questionnaire for screening of eating disorders among Mexican university students." *Eating and Weight Disorders-Studies on Anorexia, Bulimia and Obesity,* Vol. 22, No. 1, 2017, pp. 153-60.

[21] Team, R. Core. "R: A language and environment for statistical computing." 2013, p. 201.