The Relationship between Bone Mineral Density and Menstrual Cycle Abnormalities in Women with Polycystic Ovary Syndrome

Handan Çelik[1], MD, PhD,
Miğraci Tosun[2], MD, PhD, Samettin Çelik[3], MD,
Mehmet B. Çetinkaya[2], MD, PhD

1. University of Ondokuz Mayıs, School of Medicine Department of Obstetrics and Gynecology, Samsun, Turkey
2. University of Ondokuz Mayıs, School of Medicine, Department of Obstetrics and Gynecology, Samsun, Turkey.
3. Erbaa State Hospital, Department of Obstetrics and Gynecology Tokat

Correspondence:
Handan Çelik
e-mail: drhandancelik@yahoo.com

Abstract

Objective. *The aim of this study was to investigate the influence of menstrual dysfunction in the form of oligomenorrhea and amenorrhea on bone mineral density (BMD) in women with polycystic ovary syndrome (PCOS).*
Study Design. *Eighteen amenorrheic, 32 oligomenorrheic and 50 normally mensturating polycystic women participated in this study. Every woman underwent measurements of lumbar spine, femoral neck, wards, trochanter, shaft and total hip BMD by dual energy X- ray absorptiometry (DEXA).*
Results. *The difference of mean serum total testosterone levels between groups were statically significant ($p<0.01$). The bone density of the lumbar spine (L2-L4) in the group of the amenorrheic women was 0.97 ± 0.09, significantly lower than that in the eumenorrheic group (1.12 ± 0.07 SD; $p=0.001$) and the oligomenorrheic group (1.05 ± 0.05; $p=0.001$).*
Conclusions. *PCOS patients with amenorrhea have lower BMD than PCOS with eumenorrhea and oligomenorrhea. Amenorrhea is the one of the reasons of lower bone mineral density in patients with polycystic ovary syndrome.*
Keywords: *Polycystic ovary syndrome, menstrual disturbances, bone mineral density*

Introduction

Polycystic ovary syndrome (PCOS) is one of the most common endocrine disease in reproductive age women characterized by chronic anovulation and menstrual cycle disturbances like amenorrhea and oligomenorrhea, hyperandrogenemia and insulin resistance[1,2]. PCOS is seen in reproductive age group with the incidence about 5-10%[3,4,5]. Most authors agree that amenorrhea is associated with different causes and may be associated with a reduction in bone mineral density in reproductive aged women[6,7]. PCOS is a complex disorder and several components of the syndrome may affect bone density[8]. In this study, the bone

mineral density of the patient with PCOS with different menstrual status was compared.

Materials and methods

In this prospective study, 100 consecutive patients aged between 18-32 years (mean 23 years) with PCOS were studied. The study was carried out between 01 January 2007-30 April 2008 in the State Hospital, Department of Obstetrics and Gynecology, Erbaa, Turkey. An Institutional Review Board approved the study protocol, and informed consent was obtained for each patient before they were involved in the study. During the analysis of the data, all patients with PCOS were divided into three subgroups, according to the menstrual cycle length.

Group 1 - Patients with regular menstrual period

Group 2 - Patients with oligomenorrhea. Oligomenorrhea was defined as cycle length >35 days.

Group 3 - Patients with amenorrhea. Amenorrhea was defined as the absence of vaginal bleeding for 6 months.

The diagnosis of PCOS was based on according the definition of the new Rotterdam criteria[9]. The patient with PCOS was defined as having two of the following three features:

1. menstrual cycle disturbances such as oligomenorrhea and amenorrhea;
2. ultrasound features of polycystic ovaries;
3. clinical and/or biochemical features of hyperandrogenism.

Oligomenorrhea was defined by a mean cycle length exceeding 35 days. Amenorrhea was defined as the absence of vaginal bleeding for 6 months and none of women had received any medication for this condition. Vaginal ultrasound scan of the ovaries with 7.5 MHz vaginal end probe were performed in follicular phase by an experienced sonographer. Presence of ten or more sub-capsular follicles in each ovary measuring 2-9 mm in diameter and increased stroma were used to indicate the polycystic ovary (PCO) appearance. Clinical hyperandrogenism (i.e. hirsutism, acne, oily face) and biochemical hyperandrogenemia (serum total testosterone reference range: 14-76 mg/dL) were documented.

Subjects were asked to fill in a standardized questionnaire, related to their social, medical, menstrual history. All PCOS patients were in good health didn't smoke and none of them was on excessive physical training. Majority of them had similar lifestyles and diets. Serum prolactine levels and thyroid functions were measured in all participants, in any of the recruited subjects who had hyperprolactinemia or abnormal serum thyroid function tests were not included in the study. Exclusion criteria for women with PCOS also included pregnancy, cigarette-smoking, use of oral contraceptives, antidiabetics or glucocorticoids or any medication known to affect bone mineral metabolism.

The data recorded included age, parity, age at menarche, weight, height and body mass index (BMI). Body mass index was calculated as weight (kg) divided by height squared (m^2).

Laboratory measurements

Blood samples were obtained in the morning after an overnight fasting on the third day of the menstrual cycle except in those with amenorrhea. FSH, LH, E2, PRL, fasting glucose, total testosterone were all measured in all patients.

Bone Mineral Density

The BMD of the anterior-posterior lumbar spine (L2-L4), left proximal femur including the femoral neck, Ward's triangle, and the greater trochanter, shaft and total hip BMD were measured by using conventional dual- energy X-ray absorptiometry (DEXA) (LUNAR DPX-PRO; Lunar, Madison WI, USA). Technical performance was monitored by daily calibration scans using an anthropomorphic Hologic phantom.

Serum fasting glucose, HDL, LDL, total cholesterol, TG, albumin, gamma globulin levels were similar in the three groups.

Statistical Analysis

All results are expressed as mean ± SD. Student's t-test and the Mann-Whitney U-test were used for comparisons of the two groups when appropriate. Overall significance of differences between three groups was analyzed by one-way ANOVA or Kruskal-Wallis tests. Pearson correlation analysis was used for linear correlations. $P<0.05$ was considered statically significant. Statistical analyze was performed using SPSS version 15 for Windows (Statistical Package for Social Science, Chicago, IL, USA).

Results

Eighteen amenorrheic, 32 oligomenorrheic and 50 normally menstruating polycystic women participated in this study. The characteristics of the patients in the three study groups are shown in Table 1. The three groups were similar in terms of mean age, and BMI ($p>0.05$). None of the subjects in either group was in receipt of significant medication, nor were they taking hormones for contraceptives or other purposes. Serum prolactine levels and thyroid function tests were within normal range in all the subjects and there were no significant differences between three groups.

Fifty of the 100 PCOS subjects (50%) had oligomenorrhea and amenorrhea since the beginning of their menarche. PCO was diagnosed by ultrasonography 68 (68%) of 100 PCOS recipients. Hyperandrogenemia was detected clinically in 43 (43%) cases and biochemically in 73 of 100 (73%). 85 women had higher LH than FSH, elevated ratio of LH/FSH >2 was detected in 75 (75%) women.

The mean values of hormonal parameters in PCOS eumenorrheic, oligomenorrheic and amenorrheic women, were listed in Table 1. There were significant differences in plasma LH level between groups ($p<0.01$). Serum LH level was significantly higher in the amenorrheic group than in the eumenorrheic and oligomenorrheic groups ($p=0.008$; $p=0.002$ respectively). Similarly serum FSH level was higher in the amenorrheic group than in the eumenorrheic group ($p=0.007$). Although the serum level of FSH in oligomenorrheic group was lower than amenorrheic group, this was statistically insignificant ($p=0.708$). There were no statistically significant differences in serum LH and FSH levels between oligomenorrheic and eumenorrheic groups. Estradiol level didn't significantly differ in between groups ($p=0.115$). The LH/FSH ratio was found to be higher in amenorrheic groups than the other two groups.

Mean serum total testosterone levels in between groups were statistically significant ($p<0.01$). Serum level of total testosterone in amenorrheic groups was significantly higher than those in eumenorrheic ($p=0.001$) and oligomenorrheic ($p=0.001$) groups. On classifying the women with PCOS into the three subgroups regarding the menstrual

Table 1

Characteristics of three subgroups of Polycystic Ovary Syndrome Patients

	Polycystic Ovary Syndrome		
	Normally menstruating (n=50)	Oligomenorrhea (n=32)	Amenorrhea (n=18)
	Mean ± SD	Mean ± SD	Mean ± SD
Age (years)	23,10 ± 5,28	22,88 ± 5,67 NS	24,18 ± 5,13 NS
Weight (kg)	68,26 ± 18,57	64,94 ± 13,92 NS	67,76 ± 16,75 NS
Height (cm)	158,52 ± 6,13	155,63 ± 6,49 NS	157,47 ± 6,91 NS
BMI (grs/cm)	27,05 ± 4,90	27,45 ± 5,70 NS	28,48 ± 8,14 NS
Fasting Glucose (mg/dl)	96,76 ± 11,53	99,09 ± 8,74 NS	92,24 ± 9,05 NS
Total Testosterone (ng/ml)	83,23 ± 15,04	90,11 ± 15,22**	156,14 ± 41,38**
FSH (IU/l)	4,10 ± 2,46	4,58 ± 1,92**	6,26 ± 3,23**
LH (IU/l)	10,84 ± 4,41	9,72 ± 4,12**	16,28 ± 12,01**
PROLACTIN (ng/ml)	12,47 ± 7,02	15,01 ± 7,92 NS	20,56 ± 20,45 NS
E2 (pmol/l)	125,14 ± 95,45	83,63 ± 47,10 NS	84,67 ± 68,40 NS

**statistical significant p<0.05; ** statistical significant p<0.01; NS - Not statistical significant*

Table 2

BMD values for three subgroups of Polycystic Ovary Syndrome Patients

	Polycystic Ovary Syndrome		
	Normally Menstruating (n=50)	Oligomenorrheic (n=32)	Amenorrheic (n=18)
L2-L4 (gms/cm² ± SD)	1,12 ± 0,07	1,05 ± 0,05**	0,97 ± 0,09**
Femur neck (gms/cm² ± S)	0,98 ± 0,08	0,94 ± 0,11**	0,88 ± 0,11**
Wards (gms/cm² ± SD)	0,91 ± 0,11	0,84 ± 0,12**	0,79 ± 0,15**
Trochanter (gms/cm² ± SD)	0,81 ± 0,09	0,76 ± 0,09**	0,71 ± 0,14**
Shaft (gms/cm² ± SD)	1,17 ± 0,13	1,10 ± 0,12**	1,03 ± 0,15**
Total hip BMD (gms/cm² ± SD)	1,00 ± 0,09	0,94 ± 0,10**	0,88 ± 0,13**

** statistical significant p<0.05; ** statistical significant p<0.01; NS - Non statistical significant*

cycle length, it was found that there were statistically significant differences between groups (Table 2). The bone mineral density of the lumbar spine (L2-L4) in the group of the amenorrheic women was 0.97 ± 0.09 SD, and it was significantly lower than that in the eumenorrheic group (1.12 ± 0.07 SD; p=0.001) and the oligomenorrheic group (1.05 ± 0.05; p=0.001). The bone density of the lumbar spine (L2-L4) in the oligomenorrheic group was significantly lower than the eumenorrheic group (p=0.001) (Figure 1). The bone density of the femoral neck was also statistically lower in amenorrheic group (p=0.002) than in eumenorrheic and oligomenorrheic groups (Figure 2). There was no difference in between femoral density of eumenorrheic (p=0.173) and oligomenorrheic (p=0.175) groups. Similarly the wards, trochanter and the shaft BMD in amenorrheic group (p=0.001; p=0.001; p=0.001 respectively) were significantly lower than in oligomenorrheic and eumenorrheic groups. There were no statistically significant difference between oligomenorrheic and eumenorrheic groups in the wards, trochanter and the shaft BMD.

Discussion

Polycystic ovarian disease is the one of the most common cause for menstrual dysfunction[10]. In literature, among unselected women, the prevalence of PCOS was defined 7% by the presence of oligomenorrhea and biochemical hyperandrogenism[11], 4% by the presence of oligo-ovulation and clinical hyperandrogenism[12]. In this study, the prevalence of PCOS among unselected women was estimated 6% by the presence of oligo- or an-ovulation and biochemical hyperandrogenism, 5% by the presence of oligo- or a-menorrhea and

clinical hyperandrogenism. In the present study, the 2003 Rotterdam consensus workshop was used. It has expanded the diagnostic criteria for PCOS by including women with polycystic ovaries and hyperandrogenism but normal ovulatory function; and those with polycystic ovaries and normal ovulation but no clinical an biochemical findings of hyperandrogenism[9,13].

BMI, parity, duration of menstrual irregularity were not different between three groups, these findings reassure us that a selection bias with a major influence on the results had not taken place. Patients older than 35 years were excluded from the study because a decline in bone mineral density has been reported in premenopausal women[14]. The mean age was not different between PCOS groups and we didn't find correlation between age and bone mass in our study.

Women with PCOS produces estradiol lower than the normal women with normal menstrual cycle[15]. Lower estradiol production with the absence of the estradiol surge associated with ovulation, are the reasons that affect negatively the bone mineral density of women with PCOS. However production of androgens are increased in women with PCOS and chronic elevation of androgens cause positive effect on bone in women with PCOS[16].

The degree of hypoestrogenism may predict bone density measures in women with PCOS. So the serum levels of estradiol were measured on the third day of menstrual cycle, but there was no difference between groups. Because the great variability in the estradiol levels across the menstrual cycle, measuring the serum levels of estradiol only in the third day of menstruation did not help us.

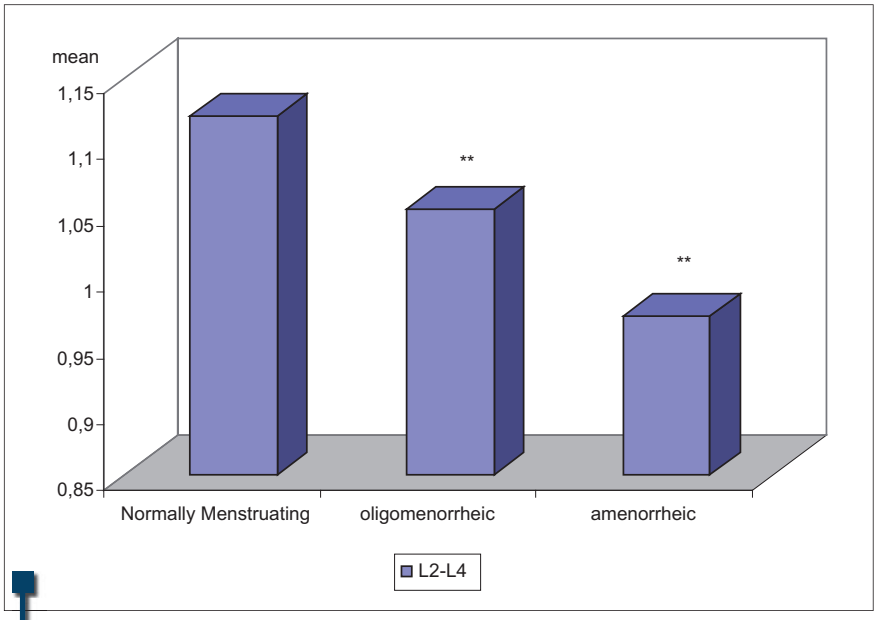

Figure 1. The bone density of the lumbar spine (L2-L4) in the oligomenorrheic and the eumenorrheic groups

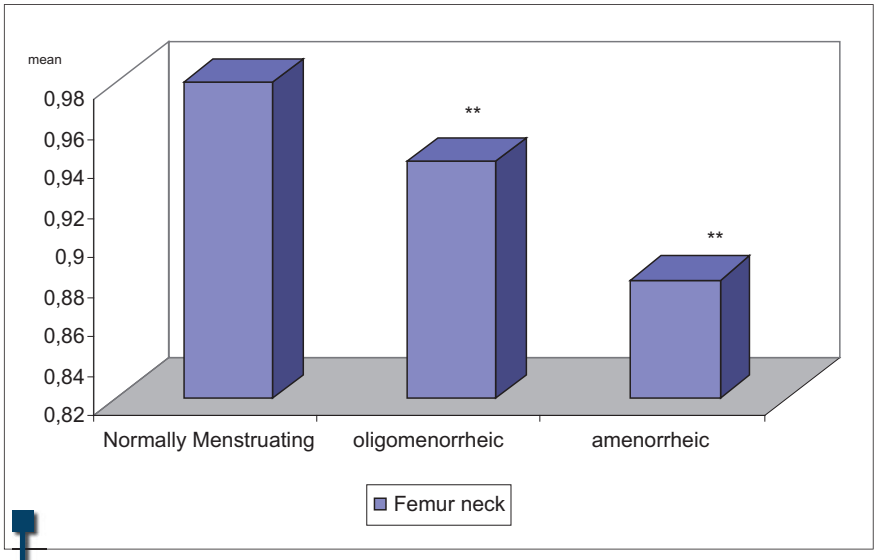

Figure 2. The bone density of the femoral neck in the amenorrheic, eumenorrheic and oligomenorrheic groups

Chronic elevation of androgens may exert a positive influence on bone in women with PCOS. The positive influence of androgens on BMD have been explained by direct (androgenic) and indirect (estrogenic) effects[17]. Direct effects of androgens occur through androgen receptors on bone related cells and indirect effects occur after conversion of androgens to 17ß-estradiol and estrone in peripheral tissues[16]. DiCarlo et al compared BMD of the amenorrheic women due to PCOS with the other reasons causing amenorrhea except from PCOS. Patient with PCOS related amenorrhea had significantly higher BMD compared to non-PCOS amenorrheic patients[18]. Recent investigations have demonstrated a potential interaction between androgen excess and menstrual irregularity in the determination of BMD in women. Prezelj et al found that hyperandrogenic but eumenorrheic women had significantly higher BMD than hyperandrogenic amenorrheic women[17]. Adami et al have evaluated endocrine profiles, BMD, bone turnover markers in 51 patients with PCOS, 26 patients with hypothalamic amenorrhea, 24 patient with idiopathic hirsutism and 35 healthy women. In the subgroup of PCOS patients with amenorrhea, spinal and femoral neck BMD were significantly lower than PCOS patients with non-amenorrhea and patient with idiopathic hirsutism[19]. In a study, Castelo-Branco et al evaluated BMD at the lumbar spine in non-obese young hirsute women and

they concluded that menstrual history was a determinant of bone density. 27 hirsute women with oligomenorrhea or amenorrhea were compared to 25 hirsute women with eumenorrhea and non-hirsute controls. Women with menstrual irregularities, both hirsute and non-hirsute had significantly lower BMD than those with regular menses[20]. In the present study, the findings were similar with literature. It was observed that amenorrheic women with PCOS had lower BMD in all sites measured than eumenorrheic and oligomenorrheic women with PCOS. However, it was demonstrated that amenorrheic women with PCOS had significantly higher serum total testosterone than oligomenorrheic and eumenorrheic women with PCOS. Therefore, it was concluded that androgens could not show their positive effect on BMD without the mid-cycle estradiol peak and/or luteal phase progesterone production in women with PCOS suggesting that estradiol plays a critical role and shows very important positive effect on bone in women with PCOS. Also it was concluded that PCOS is a complex disorder, and except from androgens, several other components of this syndrome are related with bone mass in women with PCOS.

In the present study is limited by the lack of nutritional evaluation and exact information about physical activity of study participants. In the present study, DEXA (which is the preferred method for assessing BMD) was used because of its speed, availability, and low radiation exposure especially for children. However, DEXA is limited because it measures a two dimensional area but not a volumetric BMD. Volumetric bone measurements allow assessment of bone density independent of bone size.

A family history of osteoporosis is an important factor for low BMD. The low number of women in this study reported a family history of osteoporosis or other bone-related disorders. So it didn't make a meaningful analysis of this risk factor.

This study still had several limitations. The clinical diagnosis of hyperandrogenism was made subjectively. We didn't use a standardized scoring method. The biochemical diagnosis of hyperandrogenism was made by measuring only serum levels of total testosterone. Normal reference range of androgens and the other hormonal measurements as defined by manufacturers was used without establishing normative values for our own population.

It was concluded that amenorrheic women with PCOS had lower BMD than eumenorrheic and oligomenorrheic ones. This study indicates that restoration of menstruation in amenorrheic women with PCOS will lead to improvement in bone mass.

Although serum levels of total testosterone were higher in amenorrheic group than eumenorrheic group, eumenorrheic PCOS women had higher bone mass. This suggests that hormonal periodicity and estradiol are important key factors for BMD in these patients. In conclusion, amenorrhea has deleterious effect on bone mineral density in patient with polycystic ovary syndrome. In these patients androgen overproduction has mild positive effect on bone mass but bone mass is mainly affected from hormonal periodicity and the presence of the estradiol surge associated with ovulation. ■

References

1. Cheung A, Chang R, Polycystic ovary syndrome. Clin Obstet Gynecol 1990; 33: 655-667.
2. Knochenhauer ES, Key TJ, Kahsar-Miller M, Waggoner W, Boots LR, Aziz R, Prevalence of the polycystic ovary syndrome in unselected black and white women of the southeastern United States: a prospective study. J Clin Endocrinol Metab 1998; 83: 3078-3082.
3. Diamanti-Kandarakis E, Piperi C, Genetics of polycystic ovary syndrome: searching for the way out of the labyrinth. Hum Reprod Update 2005; 11: 631-643.
4. Schroder AK, Tauchert S, Ortmann O, Diedrich K, Weiss JM, Insulin resistance in patients with polycystic ovary syndrome. Ann Med 2004; 36: 426-439.
5. Hahn S, Tan S, Elsenbruch S et al, Clinical and biochemical characterization of women with polycystic ovary syndrome in North Rhine-Westphalia. Horm Metab Res 2005: 37; 438- 444.
6. Harper KD, Weber TJ, Secondary osteoporosis Endocrinol Metab Clin North Am 1998; 27: 325-348.
7. Baird DT, AmenorrheaLancet 1997: 350; 275-279.
8. Noyan V, Yücel A, Sagsoz N, The association of bone mineral density with insulin resistance in patients with polycystic ovary syndrome. Eur J of Obstet Gynecol Rep Bio 2004; 15: 200-205.
9. The Rotterdam ESHERE/ASRM - sponsored PCOS consensus workshop group Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. Hum Reprod 2004; 9: 505-514.
10. Koivunen R, Laatikainen T, Tomas C, Huhtaniemi I, Tapanainen J, Martikainen H, The prevalence of polycystic ovaries in healthy women. Acta Obstet Gynecol Scand 1998; 78: 137-141.
11. Adams J, Polson DW and Franks S, Prevalence of polycystic ovaries in women with anovulation and idiopathic hirsutism. Br Med J (Clin Res Ed) 1986; 293: 355-359.
12. Balen A and Michelmore K, What is polycystic ovary syndrome? Are national views important? Hum Reprod 2002; 17: 2219-2227.
13. Balen AH, Laven JS, Tan SL, Dewailly D, Ultrasound assessment of the polycystic ovary: international consensus definitions. Hum Reprod Update 2003; 9: 505-514.
14. Riggs BL, Wahner HW, Melton LJ, Richelson LS, Judd HL, Offord KP, Rates of bone loss in the appendicular and axial skeletons of women: evidence of substantial vertebral bone loss before menopause. J Clin Invest 1986; 77: 1487-1491.
15. Hillard PA, Menstruation in young girls: a clinical perspective. Obstet Gynecol 2002; 99: 655-662.
16. Zborowski JV, Cauley JA, Tallbott EO, Guzick DS, Winters S, Bone mineral density, androgens and the polycystic ovary; the complex and controversial issue of androgenic influence in female bone. J Clinical Endocrinol Metab 2000; 85: 3496-3506.
17. Prezelj J, Kocijancic A, Bone mineral density in hyperandrogenic amenorrhea. Calcif Tissue Int 1993; 52: 422-424.
18. DiCarlo C, Shoham Z, MacDougall J, et al, Polycystic ovaries as arelative protective factor for bone mineral loss in young women with amenorrhea. Fertil Steril 1992; 57: 314-319.
19. Adami S, Zamberlan N, Castello R, Tosi F, Gatti D, Moghetti P, Effect of hyperandrogenism and menstrual cycle abnormalities on bone mass and bone turnover in young women. Clin Endocrinol 1998; 48: 169-173.
20. Castelo-Branco C, Pons F, Martinez de osaba MJ, et al, Menstrual history as a determinant of current bone density in young hirsute woman. Metabolism 1997, 45: 515-518.