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Effects of spironolactone on calcium and parathyroid hormone in polycystic ovary syndrome

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ABSTRACT

Aim: To investigate the effects of short-term oral intake of spironolactone on parathyroid hormone (PTH) and calcium levels, as well as other biochemical and clinical parameters in normotensive female patients with polycystic ovary syndrome (PCOS).

Method: 32 PCOS patients were studied at baseline and three months after regular intake of a daily dose of 50 mg or 100 mg spironolactone. PTH, calcium and other biochemical tests as well as clinical parameters were evaluated at baseline and after three months.

Results: Body mass index and serum PTH levels decreased significantly after treatment (*p*: 0.021 and *p*: 0.043, respectively). Mean high-density lipoprotein was significantly decreased after treatment (*p*: 0.011). No significant change was observed in calcium, phosphorus, total cholesterol, triglycerides, and fasting blood glucose levels. Serum testosterone and dehydroepiandrosterone sulphate decreased significantly after 3 months of therapy (*p*: 0.000 and *p*: 0.006; respectively).

Conclusions: In our study, taking spironolactone at a dose of 100 mg/day lowered PTH levels and had a positive effect on weight loss and insulin resistance. New studies are needed to investigate the effects of treatment with spironolactone on bone and metabolism. It is recommended to monitor dyslipidemia while taking the drug.

Key words: Polycystic ovary syndrome, spironolactone, calcium, parathormone.

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Introduction

Chronic anovulation and hyperandrogenism are characteristics of polycystic ovary syndrome (PCOS). It affects approximately 6-20% of women of reproductive age [1] Spironolactone, an androgen receptor blocker and aldosterone antagonist, is used to treat hirsutism [2]. Patients are advised of the side effects of spironolactone, which include electrolyte disturbances and volume loss [3]. There is a relationship between parathyroid hormone (PTH) and the renin-angiotensin-aldosterone system (RAAS). Aldosterone release in the zona glomerulosa of the adrenal glands is thought to be influenced by PTH [4]. This was demonstrated by Mazzocchi et al [5], who discovered that PTH stimulates the release of aldosterone from adrenocortical cells via the PTH/PTH-related peptide receptors.

Increased RAAS activity and elevated aldosterone levels have been demonstrated in patients with primary hyperparathyroidism (PA) [6]. Experimental studies in rats have shown that aldosterone excess is associated with secondary hyperparathyroidism, which is reversible by mineralocorticoid receptor antagonist therapy [7, 8]. In humans, few data are available on this topic [9, 10]. The study by Pilz at al. showed that PA is related to hyperparathyroidism and that drug or surgical treatment of PA leads to a decrease in PTH levels [9]. In another study, it was shown that the use of RAAS inhibitors is associated with a decrease in PTH levels [10]. The aim of our study is to investigate the effects of short-term oral intake of spironolactone on PTH and calcium levels as well as other biochemical and clinical parameters in normotensive female patients with PCOS.

Materials and metods

This study was conducted in Endocrinology and Internal Medicine Clinics of the University of Health Science Antalya Training and Research Hospital. The study was approved by the Ethics Committee of the University of Health Sciences, Antalya Training and Research Hospital (Date: 26/01/2023, Number: 2/6), and written informed consent was obtained from all patients/guardians. The Helsinki Declaration was followed when conducting the study. Between January 2018 and January 2023, 32 patients with PCOS were enrolled in the study. Fourteen patients (group 1) received 50 mg/day spironolactone and 18 patients received 100 mg/day spironolactone for 3 months. The subjects did not receive any other medications. Patients younger than 18 and older than 35 years, patients with diabetes mellitus, hypertension, chronic kidney disease, cancer, osteomalacia, other bone disease, thyroid or parathyroid disease, patients taking calcium or vitamin D supplements, and patients whose 25-hydroxyvitamin D (25OHD) was less than 20 ug/L were excluded.

The diagnosis of PCOS was based on the physical features of hyperandrogenism, irregulation of menstrual cycles, elevated luteinizing hormone (LH) or LH /follicle-stimulating hormone (FSH) ratio, elevated serum total testosterone (TT) levels, and no evidence of any other etiologic disease [11]. Body mass index (BMI), blood pressure (BP), mF-G (modified Ferriman–Gallwey score), and hormonal and biochemical test results, including LH, FSH, testosterone, dehydroepiandrosterone sulphate (DHEAS), prolactin, fasting blood glucose (FBG), insulin, thyroid-stimulating hormone (TSH), PTH, creatinine, and electrolytes were determined at baseline and after three months of treatment with spironolactone. Biochemical tests were analyzed spectrophotometrically using the Beckman Coulter AU5800 (Beckman Coulter Inc. CA, USA) autoanalyzer. Insulin and other necessary hormone tests were analysed by the chemiluminescence method on the Beckman Coulter DxI800 (Beckman Coulter Inc. CA, USA) analyzer. (Reference ranges; FBG: 74-106 mg/d, creatinine: 0.66-1.09 mg/dL, albumin: 35-52 g/L, calcium: 8.8-10.6 mg/dL phosphorus: 2.5-4.5 mg/dL, sodium: 136-146 mmol/L, potassium: 3.5-5.1 mmol/L, total cholesterol (TC) < 200 mg/dL, low-density lipoprotein (LDL): 10-100 mg/dL, high-density lipoprotein (HDL): 40-60 mg/dL,triglycerides (TG): 10-150 mg/dL, 25OHD: 30-100 ug/L, PTH: 12-88 ng/L, TSH: 0.34-5.6 uIU/mL, FT3 (free triiodothyronine): 2.5-3.9 ng/dL, FT4 (free thyroxine): 0.61-1.12 ng/dL, TT: 0.1-0.75 µg/L, DHEAS: 18-391 µg/dL, prolactin: 3,34-26,72 µg/L.

In a seated position, the right upper arm's BP was monitored using a standard

sphygmomanometer. Following an overnight fast, blood samples were taken in the morning. The HOMAIR (homeostasis model assessment-insulin resistance) value was calculated according to the formula (plasma glucose [mmol/L] × insulin [pU/mL]: 22.5) [12].

Statistical analysis

The SPSS (Statistical Package for the Social Sciences) 23.0 package program was used for analysis. Numerical measurements were summarized as mean ± standard deviation or median (minimum-maximum). Normality of distribution was examined using the Shapiro-Wilks test. When comparing continuous parameters between 2 groups, the independent Student t test and was used for the parameters with normal distribution, and the Mann Whitney u test was used for the parameters that did not

Table 1. Clinical characteristics and biochemical parameters of patients at baseline and after treatment with spironolactone in polycystic ovary syndrome.

Parameters (n: 32)	**Baseline**	**After treatment**	***p***
Age (years)	24.6±5.16	-	
BMI (kg/m^2)	27.8±3.2	25.9±2.6	**0.021***
mF-G score	12.4±2.2	10.2±3.1	0.062
BP (systolic) (mmHg)	124.5±10.1	120.6±10.5	0.074
BP (diastolic) (mmHg)	78.4±3.4	76.9±4.7	0.813
FBG (mg/dL)	88 (81.5-89)	89 (87-93)	0.440
Creatinin (mg/dL)	0.86±0.24	0.86±0.15	0.769
GFR (mL/min/1.73 m2)	89.21±12.69	86.12±13.50	0.321
Sodium (mmol/L)	139.41±2.81	136.55±2.29	0.072
Potassium (mmol/L)	4.34±0.21	4.61±0.77	0.095
Calcium (mg/dL)	9.57±0.35	9.37±0.24	0.065
Phosphorus (mg/dL)	3.45±0.42	3.62±0.41	0.161
25-hydroxyvitamin D (ug/L)	22.9 (20.1-24.4)	21.8 (20.1-23.7)	0.982
Magnesium (mg/dL)	2.23(1.90-2.34)	2.21 (1.92-2.38)	0.348
Albumin (g/L)	4.30±0.20	4.25±0.30	0.166
PTH (ng/L)	53 (44-64)	46 (37-54)	**0.043***
TSH (uIU/mL)	2.81±0.49	2.63±0.62	0.493
HOMAIR	2.62±1.76	2.48±1.98	0.823
TG (mg/dL)	83.81±26.63	85.81±27.55	0.712
TC (mg/dL)	181.65±38.37	182.97±31.63	0.865
HDL (mg/dL)	40.50±9.21	34.42± 6.88	**0.011***
LDL (mg/dL)	131.62±29.55	136.89±27.11	0.051
Testesterone (µg/L)	0.71±0.57	0.58±0.37	**0.000***
DHEAS (µg/dL)	319.18±135.22	289.21±136.62	**0.006***
Prolactin (µg/L)	21.43±3.40	19.20±1.23	0.198

BMI: Body mass index, m F-G (modified Ferriman–Gallwey), BP: Blood pressure, FBG: Fasting blood glucose, PTH: Parathyroid hormone, TSH: Thyroid-stimulating hormone, HDL: High-density lipoprotein, LDL: Low-density lipoprotein, TC: Total cholesterol, TG: Triglyceride, DHEAS: Dehydroepiandrosterone sulphate, HOMAIR: Homeostasis model assessment-insulin resistance, p<0.05 is statistically significant.

have normal distribution. Paired t-test (parametric distribution) and Wilcoxon test (nonparametric distribution) were used to compare the results before and after treatment. The relationship between numeric variables was analyzed using Pearson and Spearman correlation analysis. *P* <0.05 were statistically significant for all tests.

Results

The clinical characteristics and biochemical parameters of the patients at the beginning and after spironolactone treatment in polycystic ovary syndrome are presented in table 1. There were 32 patients. Treatment with spironolactone did not show clinical efficacy when used in the short term, as m F-G scores did not decrease within three months (*p*: 0.62). After treatment, 3 patients complained of fatigue, 2 of dry mouth, and 2 of polymenorrhea, and no other complications occurred. Serum PTH level decreased significantly (*p*: 0.043), while calcium level did not change after treatment (p: 0.065). HDL decreased (p: 0.011), LDL (*p*: 0.051) and TG (*p*: 0.712) did not change. DHEAS (*p*: 0.006) and TT (p: 0.000) decreased significantly after

Table 2. Clinical findings and laboratory results of polycystic ovary syndrome patients taking 50 mg/day spironolactone (group 1) and 100 mg/day spironolactone (group 2).

Parameters	Group 1 (n:14)	Group 2 (n=18)	*p*
Age (years)	23.7±4.2	24.9±5.3	0.981
BMI (kg/m^2)	27.6±3.7	27.4±3.6	0.144
mF-G score	11.9±2.3	12.8±2.4	**0.041***
BP (systolic) (mmHg)	122.6±16.1	126.2±14.2	0.073
BP (diastolic) (mmHg)	76.6±2.4	79.1±3.60	**0.033***
FBG (mg/dL)	87 (81.5-86)	86 (83-89)	0.221
Creatinine (mg/dL)	0.86±0.21	0.87±0.19	0.125
GFR (mL/min/1.73 m2)	86.22±12.79	87.14±20.50	0.365
Sodium(mmol/L)	139.44±2.86	138.52±2.28	0.266
Potassium(mmol/L)	4.32±0.22	4.38±0.71	0.801
Calcium (mg/dL)	9.57±0.31	9.44±1.20	0.080
Phosphorus (mg/dL)	3.41±0.49	3.39±0.47	0.151
25OHD (ug/L)	20.98 (20.12-21.65)	23.10 (22.24-24.65)	**0.043***
Magnesium (mg/dL)	2.16 (2.09-2.34)	2.21 (1.90-2.28)	0.724
Albumin (g/L)	4.20±0.40	4.23±0.36	0.786
PTH (ng/L)	50 (44-60)	48 (46-64)	0.588
TSH (uIU/mL)	2.82±0.48	2.33±0.79	0.321
HOMAIR	2.55±0.91	2.73±1.65	**0.011***
TG (mg/dL)	82.62±24.33	84.91±22.93	0.420
TC (mg/dL)	179.86±40.31	182.91±36.77	0.396
HDL (mg/dL)	42.53±7.61	38.95±9.86	**0.017***
LDL (mg/dL)	133.26±24.57	128.43±32.71	0.487
Testesterone (µg/L)	0.69±0.22	0.72±0.66	0.588
DHEAS (µg/dL)	324.16±130.23	315.72±139.75	0.592
Prolactin (µg/L)	19.16±3.34	24.63±1.61	0.061

BMI: Body mass index, m F-G (modified Ferriman–Gallwey), BP: Blood pressure, FBG: Fasting blood glucose, PTH: Parathyroid hormone, TSH: Thyroid-stimulating hormone, HDL: High-density lipoprotein, LDL: Low-density lipoprotein, TC: Total cholesterol, TG: Triglyceride, DHEAS: Dehydroepiandrosterone sulphate, HOMAIR: Homeostasis model assessment-insulin resistance, p<0.05 is statistically significant.

treatment with spironolactone. BMI decreased significantly after treatment (*p*: 0.021).

Clinical features and laboratory parameters of group 1 (women using 50mg/day spironolactone) and group 2 (women using 100 mg/day spironolactone) were examined. F-G score, diastolic BP, 25OHD and HOMA-IR were higher in group 2 when compared to group 1 (Table 2).

The baseline and after treatment results of group 1 and group 2 patients were examined. The PTH was significantly decreased in group 2 after treatment (*p*: 0.033) but calcium level was not changed (*p*: 0.080). HOMAIR (*p*: 0.019) and BMI (*p*: 0.036) were decreased significantly in group 2 after treatment. PTH and HOMAIR were not changed in group 1 after treatment (p: 0.072 and p: 0.724 respectively). While LDL increased significantly after treatment in group 2 (*p*: 0.001),

Table 3. Clinical findings and laboratory results of polycystic ovary syndrome patients taking 50 mg/day spironolactone (group 1) and 100 mg/day spironolactone (group 2) at baseline and after treatment.

Parameters	Group 1 (n:14)			Group 2 (n=18)		
	Baseline	After treatment	*p*	Baseline	After treatment	*p*
Age (years)	23.7±4.2	-	-	24.9±5.3	-	-
BMI (kg/m^2)	27.6±3.7	26.9±1.7	0.263	27.4±3.6	25.4±2.1	**0.036***
mF-G score	11.9±2.3	10.6±2.1	0.961	12.8±2.4	10.7±1.5	0.391
BP (systolic) (mmHg)	122.6±16.1	119.1±9.6	0.122	126.2±14.2	119.6±10.5	0.563
BP (diastolic) (mmHg)	76.6±2.4	75.0±2.7	0.788	79.1±3.60	77.8±2.13	0.894
FBG (mg/dL)	87 (81.5-86)	89 (87-91)	0.849	86 (83-89)	90(89-93)	0.514
Creatinine(mg/dL)	0.86±0.21	0.90±0.43	0.224	0.87±0.19	0.85±0.94	0.722
GFR (mL/min/1.73 m2)	86.22±12.79	83.77±13.2	0.091	87.14±20.50	84.13±11.33	0.361
Sodium (mmol/L)	139.44±2.86	138±2.60	0.920	138.52±2.28	134.2±1.82	**0.039***
Potassium(mmol/L)	4.32±0.22	4.64±0.56	**0.031***	4.38±0.71	4.55±0.26	0.062
Calcium (mg/dL)	9.57±0.31	9.36±1.50	0.170	9.44±1.20	9.37±1.82	0.080
Phosphorus (mg/dL)	3.41±0.49	3.57±0.63	0.341	3.39±0.47	3.67±0.78	0.867
25OHD (ug/L)	20.98 (20.12-21.65)	21.16 (20.03-22.66)	0.820	23.10 (22.24-24.65)	20.36 (20.11-21.79)	0.945
Magnesium (mg/dL)	2.16 (2.09-2.34)	2.19 (2.11-2.27)	0.312	2.21 (1.90-2.28)	2.11(2.02-2.24)	0.224
Albumin (g/L)	4.20±0.40	4.22±1.21	0.809	4.23±0.36	4.36±1.73	0.162
PTH (ng/L)	50 (44-60)	46 (41-54)	0.072	48 (46-64)	40 (37-44)	**0.033***
TSH (uIU/mL)	2.82±0.48	2.66±0.26	0.513	2.33±0.79	2.45±1.17	0.441
HOMAIR	2.55±0.91	2.46±0.73	0.724	2.73±1.65	2.48±1.82	**0,019 ***
TG (mg/dL)	82.62±24.33	84.71±25.56	0.514	84.91±22.93	86.17±28.64	0.852
TC (mg/dL)	179.86±40.31	181.13±29.17	0.812	182.91±36.77	183.23±30.82	0.776
HDL (mg/dL)	42.53±7.61	36.23±7.13	**0.031***	38.95±9.86	32.74±6.77	**0.009***
LDL (mg/dL)	133.26±24.57	135.95±26.23	0.071	128.43±32.71	137.24±28.87	**0.001***
Testesterone (µg/L)	0.69±0.22	0.56±0.47	**0.000***	0.72±0.66	0.60±0.81	**0.000***
DHEAS (µg/dL)	324.16±130.23	295.23±131.04	**0.002***	315.72±139.75	282.93±140.96	**0.005***
Prolactin (µg/L)	19.16±3.34	18.95±1.41	0.227	24.63±1.61	20.77±3.61	0.077

BMI: Body mass index, m F-G (modified Ferriman–Gallwey), BP: Blood pressure, FBG: Fasting blood glucose, PTH: Parathyroid hormone, TSH: Thyroid-stimulating hormone, HDL: High-density lipoprotein, LDL: Low-density lipoprotein, TC: Total cholesterol, TG: Triglyceride, DHEAS: Dehydroepiandrosterone sulphate, HOMAIR: Homeostasis model assessment-insulin resistance, p<0.05 is statistically significant.

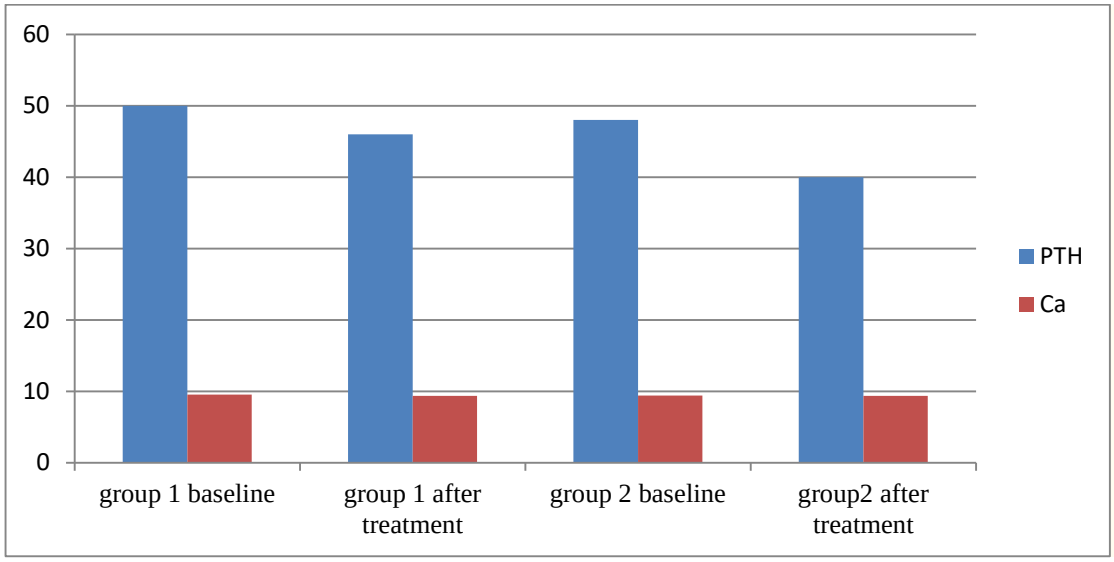

Figure 1. The graph shows the data of parathyroid hormone and calcium from baseline and after treatment of two groups.

no significant difference was observed in group 1 (*p*: 0.071) after treatment (Table 3, Figure 1).

Discussion

There are studies in the literature that indicate a relationship between aldosterone and PTH secretion [9,10]. In both animal and human studies, it has been observed that urinary calcium excretion increases after high sodium intake or mineralocorticoid intake and that PTH increases secondary to a decrease in serum calcium [13-16]. Rossi et al. found that PTH levels were significantly higher and ionized calcium lower in 10 patients with PA compared with healthy controls, and they observed that PTH levels decreased and calcium levels increased in this group of patients after taking spironolactone [13]. In another study, Rossi et al. showed that urinary calcium excretion increased under saline, and this increase was higher in the PA group than in the essential hypertension group. They also observed that PTH levels were more stimulated in the PA than in the essential hypertension [14]. In animal studies investigating the effects of spironolactone on bone, aldosterone was found to cause a decrease in serum calcium level with calciuresis and a decrease in bone mineral density with a secondary PTH increase, and this situation can be reduced by spironolactone [15,16]. In their study of patients with PA, Adolf C. et al. found that urinary calcium excretion decreased after surgery or treatment with spironolactone, and they observed a decrease in bone turnover markers in the group treated with spironolactone, highlighting the bone-protective effect of spironolactone [17]. Our study showed that in normotensive women spironolactone had no effect on PTH at a dose of 50 mg/day but at a dose of 100 mg/day a decrease in PTH levels were observed. No effect on calcium was detected. This limited number of studies in the literature supports that spironolactone may have an effect on PTH. However it is known that various environmental factors including diet, exercise, smoking, BMI and alcohol can also influence PTH levels [18]. In three larger studies involving more participants, it was found that smoking has a lowering effect on PTH levels [19-21]. These studies also show that PTH increases with increasing BMI and there is a positive correlation between PTH and BMI [19-21]. Since

BMI also decreased in our patient group receiving 100 mg spironolactone, it is difficult to say whether the change in PTH is due to spironolactone or the weight change in the patients. Study results are conflicting regarding the effects of spironolactone on BMI. In the study by Long et al, significant weight loss and a significant decrease in HOMA-IR were observed in PCOS patients, both in the group receiving spironolactone alone and in the group receiving metformin [22]. However no change in weight was observed in the study by Nakhjavani et al [23]. In our study, the group of patients taking 100 mg spironolactone, there was also a significant decrease in BMI and HOMAIR scores after treatment compared to baseline. Since the follow-up period of our study was short, it is difficult to say whether the weight loss in our study was due to the patient's diet and exercise or the effect of the spironolactone. Spironolactone is a drug that is very beneficial in the treatment of PCOS, especially with regard to hirsutism [24,25]. The effectiveness of spironolactone in the treatment of hirsutism is due to its ability to compete with dihydrotestosterone at the androgen receptor. In addition, spironolactone blocks cytochrome P450 enzymes in the ovaries and adrenal gland and lowers androgen levels [26]. Our study shows that both 50 mg/day and 100 mg/day of spironolactone lower androgen levels. The absence of a decrease in m F-G score is likely due to the short duration of treatment. These results suggest that spironolactone may have effects on weight and insulin resistance in PCOS in addition to its antiandrogenic effects. In studies on the effect of spironolactone on lipids different results have been obtained [23,27,28]. Gökmen et al [28] and Nakhjavani et al [23] found a decrease in HDL levels when spironolactone was used in patients with hirsutism, similar to our study.

In our study, the duration of spironolactone use was short and the number of patients was small, which are the main limitations of our study. Since the follow-up period of the patients was short and no repeated PTH measurements could be performed, long-term changes in PTH levels could not be observed. As markers of bone formation and resorption and bone densitometry were not evaluated, the effect of spironolactone on osteoporosis could not be investigated.

Conclusions

Short-term use of spironolactone did not produce clinical benefit in hirsutism, although it lowered androgen levels. A decrease was observed in PTH levels after 3 months use of 100 mg/day spironolactone. Since PTH can be influenced by many environmental factors, it is difficult to say that the change in PTH level in our cohort is directly related to spironolactone or not. A prospective study with a large number of patients could increase the significance of the study and support the effects of spironolactone on PTH in the future. The observed weight loss after treatment needs to be supported by further studies, and it is recommended that dyslipidemia also be monitored during treatment.

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