ORIGINAL STUDY

Взаимосвязь маркеров ремоделирования костной ткани с минеральной плотностью костей у женщин с сахарным диабетом 2 типа, находящихся в постменопаузе

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Цель. *Определить связи маркеров костного ремоделирования с минеральной плотностью костной ткани (МПК), метаболическими параметрами и композитным составом тела (КСТ) у женщин с сахарным диабетом 2 типа (СД2), находящихся в постменопаузе.*

Материалы и методы. *В исследование включено 140 женщин с длительностью СД2 более 5 лет. Контрольную группу составили 20 женщин в постменопаузе, без СД, с нормальной МПК. Исследование МПК и КСТ осуществляли с помощью двухэнергетической рентгеновской абсорбциометрии. На основании Т-критерия больные СД2 разделены на группы с нормальной МПК (n=50), остеопенией (n=50) и остеопорозом (n=40). Концентрацию в сыворотке крови маркеров костеобразования остеокальцина и С-терминального пропептида коллагена I типа (CICP), ингибитора костной резорбции остеопротегерина, паратгормона, а также мочевую экскрецию фрагментов С-терминальных телопептидов коллагена I типа (alphaCrossLaps, или CTX-I; маркер костной резорбции) определяли с помощью ИФА.*

Результаты. *Уровень остеокальцина был снижен у женщин с нормальной МПК, остеопенией и остеопорозом ($p<0,0002$), различий между группами больных с разной МПК не выявлено. Концентрация остеопротегерина была снижена во всех группах больных, у больных с остеопенией и остеопорозом уровень маркера был ниже, чем у пациентов с нормальной МПК ($p=0,003$ и $p=0,01$). У женщин с остеопорозом экскреция CTX-I была выше, чем в контроле и у больных с нормальной МПК ($p=0,01$ и $p=0,01$). Концентрация CICP не различалась между группами. Уровень паратгормона был повышен во всех группах женщин с СД2 ($p\leq0,0005$), различий в зависимости от МПК не выявлено. В многофакторном регрессионном анализе факторами, ассоциированными с МПК в позвоночнике, оказались: ИМТ, возраст и CTX-I ($R^2=0,38$, $p=0,0007$). Возраст, ИМТ, остеопротегерин и CTX-I были ассоциированы с МПК в проксимальном отделе бедра ($R^2=0,44$, $p=0,00003$). Не выявлено взаимосвязи между маркерами костного ремоделирования и уровнем HbA_{1c}, показателями липидного обмена и параметрами КСТ.*

Выводы. *Остеопороз у женщин с СД2, находящихся в постменопаузе, ассоциирован со снижением уровня циркулирующего остеопротегерина и повышением мочевой экскреции CTX-I. Полученные данные не подтверждают гипотезу о связи маркеров костного ремоделирования с метаболическими параметрами и КСТ.*

Ключевые слова: *сахарный диабет 2 типа; остеопороз; постменопауза; минеральная плотность костной ткани, маркер ремоделирования костей; композитный состав тела*

The relationships between bone turnover markers and bone mineral density in postmenopausal type 2 diabetic women

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Aim. *To determine the relationships between bone remodelling markers and bone mineral density (BMD), metabolic parameters and total body composition (TBC) in postmenopausal women with type 2 diabetes (T2D).*

Materials and methods. *The study included 140 women who were diagnosed with T2D more than five years prior. The control group included 20 postmenopausal nondiabetic women with normal BMD. The BMD and TBC parameters were assessed by dual X-ray absorptiometry. Based on their T-scores, T2D women were divided into the following groups: normal BMD (n = 50), osteopenia (n = 50) and osteoporosis (n = 40). Serum levels of bone formation markers [osteocalcin and type 1 C-terminal collagen propeptide (CICP), osteoprotegerin (an inhibitor of bone resorption), parathyroid hormone (PHT) and urinary excretion of C-terminal telopeptides of type 1 collagen (alpha-CrossLaps, or CTX-I; a bone resorption marker)] were determined by ELISA.*

Results. *Osteocalcin levels were decreased in all groups of T2D women (all $P < 0.0002$), without any differences between groups. Osteoprotegerin levels were reduced in all patient groups but was significantly lower in diabetic women with osteoporosis and osteopenia compared to those with normal BMD ($P = 0.003$ and $P = 0.01$, respectively). Women with osteoporosis had higher urinary CTX-I excretion than control and diabetic women with normal BMD ($P = 0.01$ and $P = 0.01$, respectively).*

Received: 24.08.2016. Accepted: 12.10.2016.

Сахарный диабет. 2016;19(5):375-382 doi: 10.14341/DM8008 Diabetes Mellitus. 2016;19(5):375-382

CICP levels did not differ between groups. PHT concentrations were increased in diabetic women (P < 0.0001), without any differences between groups. After multiple regression analysis, BMI, age and CTX-I excretion were all associated with lumbar BMD (R2 = 0.38, P = 0.0007), whereas age, BMI, osteoprotegerin levels and CTX-I excretion were all predictive of BMD at the proximal femur (R2 = 0.44, P = 0.00003). There was no relationship between bone remodelling markers and HbA1c, lipid metabolism or TBC.

***Conclusions**. In postmenopausal T2D women, osteoporosis is associated with decreased serum osteoprotegerin levels and enhanced urinary CTX-I excretion. The data do not support the existence of an interrelationship between bone remodelling markers, metabolic parameters and TBC in postmenopausal women with T2D.*

***Keywords**: type 2 diabetes; osteoporosis; postmenopause; bone mineral density, bone turnover marker; body composition*

Type 2 diabetes mellitus (T2DM) and osteoporosis are two non-infectious diseases that have reached epidemic levels among the ageing population in industrialised countries. Patients with T2DM have an increased risk of fractures of the spine and hip, even in case of normal bone mineral density (BMD) [1, 2]. One of the causes of the risk of fracture despite a normal BMD may be obesity, which is associated with an increased BMD [3] and simultaneously is a risk factor for fractures [4]. Increased bone fragility in patients with T2DM may be due not only to a decrease in BMD but also due to the impairment of the bone structure (diminished bone quality) and changes in the bone remodelling process as well [5].

Previous studies have demonstrated changes in the concentrations of osteogenesis and bone resorption markers in patients with T2DM. A number of studies have shown a decrease in the blood levels of bone formation markers, such as osteocalcin [6–8] and N-terminal propeptide of collagen type I [8, 9], as well as a bone resorption marker, C-terminal telopeptide of type I collagen (CTX) [7, 9]. However, data on a link between markers of bone remodelling and BMD are fragmentary and do not show the whole picture.

The relationships between the levels of bone remodelling markers and diabetes-specific metabolic disorders are of particular interest taking into account the endocrine interactions between bones, muscles, adipose tissue and pancreatic islets [10]. The relationships between bone remodelling markers and body composition, i.e. the fat, lean and bone mass, remain to be clarified.

Aim

The aim of this study was to determine the relationships between bone remodeling markers, BMD, metabolic parameters and total body composition (TBC) in postmenopausal women with T2DM.

Material and Methods

A cross-sectional observational study was conducted. Inclusion criteria were female sex, age of 50–70 years, diagnosis of T2DM established at least five years previously and duration of menopause of no less than one year. Exclusion criteria were medical history of endocrine diseases (hypercortisolism, hyperthyroidism, hypopituitarism or polyglandular syndromes), rheumatic diseases (rheumatoid arthritis, ankylosing spondylitis, or other), digestive system diseases accompanied by an impairment in calcium absorption (malabsorbtion syndromes, bariatric surgery, etc.), non-diabetic kidney disease, or stage 4-5 chronic kidney disease (CKD) or history of a kidney transplantation, hematologic diseases, of the use glucocorticoids, immunosuppressants, thiazolidinediones, SGLT2 inhibitors, bisphosphonates, calcitonin, strontium, or postmenopausal hormonal replacement therapy.

This study was approved by the local Ethics Committee of the Scientific Institute of Clinical and Experimental Lymphology in Novosibirsk, Russia (minute no. 104, 20.10.2014). All patients gave informed consent to participate in the study.

This study included 140 women with T2DM, between 50 and 70 years of age (median 63 years). The duration of menopause varied from 1 to 39 years (median 14 years). Eleven patients had normal weight, 38 were overweight and 91 were obese (including 39 subjects with class I obesity, 35 ones with class II and 17 patients with class 3). The body mass index (BMI) range was 21.7–50.3 kg/m2 (median, 32.5 kg/m2).

The duration of diabetes from the time of diagnosis varied from 5 to 42 years (median 15 years). Most study participants received insulin therapy (n = 122). In addition to insulin, 64 patients took metformin, 5 patients took sulfonylureas, 16 ones received combination of metformin and sulfonylurea and 37 received insulin monotherapy. Ten patients received metformin monotherapy, and 8 took metformin in combination with sulfonylureas. Complications of T2DM included neuropathy (n = 138), retinopathy (n = 118) and peripheral artery disease (n = 77). Stage 2 CKD was verified in 51 women, 46 ones had CKD 3a and five women had CKD 3b.

Comorbidities included hypertension (n = 137) and coronary artery disease (n = 65). Four patients reported smoking. Lipid-lowering therapy with statins was given to 70 patients; 4 received therapy with fibrates, and 5 had a statin–fibrates therapy.

The control group included 20 postmenopausal women between 50 and 70 years of age with normal glucose tolerance and normal BMD. The inclusion and exclusion criteria were the same as those for the study group.

The assessment of BMD was conducted using a dual-energy X-ray absorptiometry Lunar Prodigy, GE, USA. The values of BMD and and T-score were assessed in the spine

(lumbar vertebrae, L1–L4), proximal femur, femoral neck and forearm of the non-dominant hand. Body composition was estimated using the "Total Body Composition" software according to the densitometer manufacturer's instructions. Bone mass, fat and lean mass and truncal mass were measured. To evaluate the distribution of adipose tissue, fat mass was measured in the central abdominal and thigh areas. Android fat mass was measured in the appropriate area, with the lower boundary at the pelvis cut and the upper boundary at 20% of the distance between the pelvis and neck and by arm lines on the sides. Gynoid fat was measured at a distance that was 1.5 times the height of the android area below the pelvis cut and by the leg line on the sides.

The risk of low-energy fractures was evaluated using a FRAX questionnaire (web version 3.8, calculated according to the Russian model). The general risk of low-energy fractures (FRAX total) and hip fracture risk (FRAX hip) were assessed.

The levels of the bone formation markers osteocalcin and C-terminal propeptide of collagen type I (CICP) and the concentrations of osteoprotegerin, a bone-resorption inhibitor, and parathyroid hormone (PTH) were determined in fasting blood samples taken at 8 a.m. Bone resorption was evaluated by the excretion of fragments of CTX-I in urine. ELISA was used for the analysis of these factors. Total osteocalcin and CTX-I alpha-CrossLaps® levels were determined by commercial kits (Immunodiagnostic Systems, UK). Osteoprotegerin levels were measured with Bender Medsystems (Austria) kits. The CICP levels determined using QUIDEL (USA) test systems. For PTH level testing, DIAsource ImmunoAssays S.A (Belgium) test systems were used. Testing was conducted according to manufacturers' instructions. The results CTX-I concentration were adjusted to the level excreted creatinine.

Statistical analysis was performed using STATISTICA 10 software (StatSoft Inc, 2011, USA). Taking into account that the distribution of most studied indicators was not normal, non-parametric statistical testing was used. Differences between groups were assessed using the Mann–Whitney U test (for two groups) and the Kruskal–Wallis ANOVA test (for three or more groups). The relationships between parameters were studied using Spearman's rank correlation analysis, multivariate stepwise regression analysis and multivariate discriminant analysis. For multivariate analysis models, logarithms of variables with non-normal distribution were used. The critical level of statistical significance was set at $P = 0.05$ when testing hypotheses. Data are presented as medians (25th and 75th percentiles).

Results

Patients were divided into groups on the basis of the lowest T-score value: normal BMD (n = 50), osteopenia (n = 50) and osteoporosis (n = 40). As shown in Table 1, women with osteoporosis, as compared to those with normal BMD, were slightly older and had a greater duration of menopause, shorter height, and lower weight and BMI. Patients with osteopenia demonstrated defferences in age, body weight and BMI from women with a normal BMD. The waist-to-hip ratio (WHR), T2DM duration, lipid metabolism parameters, glomerular filtration rate (GFR) and albumin-to-creatinine ratio (ACR) in urine did not significantly differ among the groups. The average

Table 1

Clinical and laboratory characteristics of groups

Parameter	Group of patients with T2DM		
	Normal BMD (n = 50)	Osteopenia (n = 50)	Osteoporosis (n = 40)
Age, years	60 (56; 64)	63 (59; 66)*	66 (61.5; 67)*
Duration of postmenopausal period, years	13 (7; 18)	14 (9; 22)	16 (12; 21)*
Height, cm	160 (157; 164)	158 (152; 162)	156.5 (151; 159)*
Body weight, kg	90.5 (79; 100)	78 (73; 95)*	75 (69.5; 86.5)*
BMI, kg/m^2	35.2 (31.2; 37.8)	32 (29.3; 36.3)*	31.4 (28.9; 35.3)*
WHR	1.0 (0.9; 1.0)	0.9 (0.9; 1.0)	0.9 (0.8; 1.0)
Duration of T2DM, years	15 (12; 20)	17 (12; 19)	15 (10; 20)
HbA1c, %	9.8 (8.2; 10.5)	8.7 (7.6; 9.8)*	7.8 (7.1; 9.1)*
Total cholesterol, mmol/l	5.1 (4.6; 6.2)	5.0 (4.6; 5.8)	5.1 (4.5; 5.8)
Triglycerides, mmol/l	2.1 (1.3; 2.6)	1.6 (1.1; 2.1)*	1.7 (1.2; 2.4)
LDL, mmol/l	3.5 (2.4; 4.2)	3.0 (2.2; 3.6)	3.2 (2.8; 3.7)
HDL, mmol/l	1.2 (1.1; 1.4)	1.4 (1.2; 1.6)*	1.3 (1.0; 1.5)
GFR, ml/min/1.73 m^2	64 (56; 72)	61 (49; 79)	71 (57; 85)
ACR, mg/g creatinine	14.8 (3.5; 35.4)	3.1 (1.4; 14.3)*	3.9 (1.7; 14.4)
FRAX total	6.2 (5.7; 6.8)	8.1 (7.3; 9.6)*	9.2 (8; 11)**
FRAX hip	0.1 (0.1; 0.2)	0.6 (0.4; 1)*	0.9 (0.5; 1.8)**

Notes: Data are presented as medians (25th; 75th percentiles); asterisk *: statistically significant ($p < 0.05$) difference; one *: with a group of patients with a normal BMD, two **: with a group of patients with a normal BMD and osteopenia; GFR is calculated by the formula of CKD–EPI, 2009 or by endogenous creatinine clearance test (in patients with morbid obesity).

Table 2

Serum concentrations of bone remodelling markers in women with T2DM relative to BMD

Parameter	Groups			
	Control group (n = 20)	T2DM, normal BMD (n = 50)	T2DM, osteopenia (n = 50)	T2DM, osteoporosis (n = 40)
Osteocalcin, ng/ml	16.6 (12.5; 21.7)	8.5 (6.9; 11.8)*	9.1 (6.8; 13.4)*	10,4 (5.9; 13.9)*
CICP, ng/ml	61.8 (57.6; 67.8)	64.2 (57.6; 72)	64.8 (60; 72)	64.8 (58.8; 75)
Osteoprotegerin, pg/ml	16.9 (12.9; 25)	11.2 (7.4; 17.4)*	6.6 (1.8;13.8)**	6.6 (1.8; 14)**
CTX-I, ng/g creatinin	1.02 (0.61;1.89)	1.43 (0.86; 1.93)	1.47 (1.11; 2.03)*	1,66 (1.25; 2.19)**
PTH, pg/ml	47.8 (37.8; 58.2)	74 (57; 197)	62,5 (54; 79.6)	70 (56.3; 90)

Notes: Data are presented as medians (25th; 75th percentiles); asterisk *: statistically significant ($p < 0.05$) difference; one *: with the control group; two **: with the control group and with the group of patients with a normal BMD.

level of glycated haemoglobin A1c (HbA1c) was lower in the groups of patients with osteopenia and osteoporosis. The 10-year risk of major low-energy fractures and hip fractures, according to FRAX results, was, as expected, higher with osteoporosis and osteopenia than in those and with a normal BMD (both differences are statistically significant: $p < 0.0001$).

Concentrations of osteocalcin and osteoprotegerin were significantly lower in diabetic patients as compared to control [9 (6.8; 13.2) and 16.6 (12.5; 21.7) ng/ml; $p < 0.00001$ versus 9.2 (3.8; 16.4) and 19.6 (12.9; 25) pg/ml; $p = 0.0007$]. There was no significant difference between the patients and the control group in CICP level [64.8 (58.2; 72) and 61.8 (57.6; 67.8) ng/ml; $p = 0.31$]. Urinary concentrations of CTX-I and PTH were significantly higher in patients with T2DM than in the control group [1.5 (1.08; 2.04) and 1.02 (0.61; 1.89) ng/g creatinine, $p = 0.04$; 67.1 (54; 90.7) and 47.8 (37.8; 58.2) pg/ml, $p < 0.0001$].

The levels of bone remodelling markers in patient groups with different BMDs are presented in Table 2. Osteocalcin concentrations were lower in patients with a normal BMD ($p < 0.00001$), osteopenia ($p = 0.0002$) and osteoporosis ($p = 0.0002$) than those in the control group, but there were no differences between the groups of patients with different BMDs. Osteoprotegerin concentrations were decreased in patients with normal BMD, osteopenia and osteoporosis as compared to control ($p = 0.03$, $p = 0.0005$ and $p = 0.0005$ respectively). At the same time, patients with osteopenia and osteoporosis demonstrated lower concentration of the marker when compared to normal BMD patients ($p = 0.003$ and $p = 0.01$, respectively). The excretion of CTX-I in patients with a normal BMD did not exceed the level seen in controls ($p = 0.2$); in patients with osteopenia, there was a tendency for elevation ($p = 0.06$), and only in patients with osteoporosis was there a significant increase in this parameter ($p = 0.01$). In patients with osteoporosis, CTX-I levels were significantly higher than in women with a normal BMD ($p = 0.04$). We found no differences in concentrations of CICP between the groups. The levels of PTH were elevated in all patient groups when compared to control ($p \leq 0.0005$ for all groups), but there were no differences between groups with different BMD ($p \geq 0.11$).

Rank correlation analysis (Table 3) showed a positive relationship between osteoprotegerin levels and BMD in the spine ($r = 0.18$; $p = 0.03$), proximal femur ($r = 0.23$; $p = 0.006$) and femoral neck ($r = 0.3$; $p = 0.0003$). There was a reverse correlation between CTX-I excretion and BMD and T-score in the spine ($r = -0.19$; $p = 0.02$). There were no correlations between the levels of osteocalcin, CICP and PTH and densitometry parameters.

Considering the results of the group comparisons and correlation analysis, multivariate stepwise regression analysis included age, BMI, duration of menopause and levels of osteoprotegerin and CTX-I as independent variables and BMD in the spine, proximal femur and forearm as dependent ones. The age and CTX-I levels were associated with lumbar BMD in a multiple regression ($\beta = 0.213$, $\beta = -0.23$ and $\beta = -0.19$, respectively, $R2 = 0.38$; $p = 0.0007$). In the proximal femur, factors associated with BMD were age, BMI and osteoprotegerin and CTX-I levels ($\beta = -0.3$, $\beta = 0.198$, $\beta = 0.208$ and $\beta = -0.17$, respectively, $R2 = 0.44$; $p = 0.00003$). In the forearm, factors associated with BMD were age and BMI ($\beta = -0.38$ and $\beta = 0.245$, respectively, $R2 = 0.43$; $p = 0.0006$).

Table 3

Correlation between the concentrations of bone remodelling markers and BMDs in various parts of the skeleton in women with type 2 diabetes mellitus

Bone remodelling factor	BMD			
	Spine	Proximal femur	Femoral neck	Forearm
Osteocalcin	-0.04	-0.02	0.02	-0.1
CICP	0.01	-0.03	-0.07	0.07
Osteoprotegerin	0.18*	0.23*	0.3*	-0.09
CTX-I	-0.19*	-0.15	-0.14	-0.12
PTH	0.04	0.07	0.12	0.01

Table 4

Osteoporosis predictors in multivariate analysis in women with T2DM

Combination of predictors	Model parameters	
	Recognition quality, %	p
Age + BMI	72.4	<0.0001
Age + BMI + osteoprotegerin* + CTX-I*	74.8	<0.006
Age + BMI + duration of postmenopause	84.6	<0.0009
Age + BMI + duration of postmenopause + osteoprotegerin* + CTX-I*	75	<0.0014

*Logarithms of variables were used

In the multivariate discriminant analysis (Table 4), the combination of age, BMI and duration of menopause was the most informative predictor of osteoporosis in women with T2DM (classification accuracy, 84.6%). The inclusion of bone remodelling parameters, such as osteoprotegerin and CTX-I, which were linked to BMD in other models of statistical analysis, did not improve the quality of osteoporosis prediction.

The osteoprotegerin level showed a weak negative correlation with the 10-year risk of osteoporotic fractures (FRAX total r = -0.22; p = 0.009) and with the risk of hip fracture (FRAX hip r = –0.22; p = 0.009). There were no correlations between other bone remodelling markers and FRAX indices.

No correlation was established between the concentrations of bone remodelling markers and HbA1c, lipid metabolism parameters, BMI or WHR. The PTH level demonstrated a positive correlation with HbA1c (r = 0.21; p = 0.01) and triglycerides (r = 0.25; p = 0.003) and a negative correlation with the HDL concentration (r = -0.33; p = 0.0007). Age, duration of T2DM and duration of menopause shown no correlation with the bone remodelling markers.

The osteocalcin concentration correlated weakly with ACR (r = 0.3, p = 0.01) and CFR (r = -0.19, p = 0.02). Patients with a GFR <60 ml/min/1.73 m2 tended to have higher concentrations of osteocalcin than those with a higher GFR [8.2 (6.4; 11.6) and 9.1 (7.2; 14.7) ng/ml, respectively; p = 0.08]. The levels of PTH correlated with ACR correlated with ACR (r = 0.31; p = 0.008). Other parameters of bone remodelling were not associated with CKD involvement.

Patients with osteoporosis and osteopenia, as compared to those with normal BMD, had greater total and truncal fat mass (Table 5). Women with osteoporosis had less gynoid and android fat mass compared with those with normal BMD. There was no difference in the ratio of fat mass in the central abdomen and thigh areas between patients from different BMD groups. Women with osteoporosis had less lean mass compared to those with a normal BMD.

There were no correlations between body composition parameters and bone remodelling marker or PTH levels (all $p < 0.12$ and $p > 0.4$).

Discussion

In this study, we have revealed the changes in concentrations of bone remodelling markers in postmenopausal women with T2DM. The studied markers (osteocalcin, CICP, osteoprotegerin and CTX-I) were associated differently with BMD.

We found that in patients with T2DM the level of osteocalcin, non-collagenuos protein of bone matrix synthesized by osteoblasts, is decreased, which is consistent with the results of other studies [8, 11] and the results of a meta-analysis [12]. The concentrations of CICP, which reflects the level of collagen type I synthesis, were not different from control. The causes selective deficit of osteocalcin in T2DM are unclear. It was shown previously that hyperglycemia may inhibit the synthesis of osteocalcin. In pre- or postmenopausal women, the oral glucose load reduces the blood level of non-decarboxylated osteocalcin. A supraphysiological level of glucose suppresses osteocalcin gene expression in an osteoblast culture [13]. It is possible

Table 5

Body composition in type 2 diabetic women with different BMD

Parameter	Patient group		
	Normal BMD (n = 50)	Osteopenia (n = 50)	Osteoporosis (n = 40)
Total fat, kg	36.8 (31.9; 43.2)	32.1 (27.4; 41.5)*	31.3 (25.9; 37.4)*
Truncal fat mass, kg	22.9 (20; 25.3)	19.7 (17.1; 22.9)*	18.7 (15.7; 23.4)*
Android fat, kg	4.0 (3.5; 5)	3.6 (2.9; 4.5)	3.4 (2.8; 4.5)*
Gynoid fat, kg	5.8 (4.6; 6.6)	4.8 (3.7; 6.7)	4.2 (3.4; 5.6)*
Android/gynoid fat ratio	1.14 (1.04; 1.28)	1.12 (1.06; 1.3)	1.11 (1.03; 1.31)
Lean mass, kg	47.6 (43.6; 53)	45.8 (42.8; 49.2)	42 (39; 45.8)**

Note: The asterisk * indicates statistically significant (p < 0.05) differences. One *: with a group of patients with a normal BMD, two **: with groups of patients with a normal BMD and osteopenia.

that reduced serum osteocalcin concentration in the presence of a normal CICP level serves as an indicator of osteoblast dysfunction in patients with T2DM.

We did not find differences in the concentration of osteocalcin between groups of women with different BMD. Osteoporosis in women with T2DM was associated with a decreased blood level of osteoprotegerin, an inhibitor of bone resorption, and with increased urinary excretion of the bone resorption marker CTX-I. It was experimentally shown that a high level of glucose reduces the expression of osteoprotegerin in an osteoblast culture [14]. A reduced level of osteoprotegerin synthesis in patients with T2DM may promote the activation of bone resorption. We have recorded an increase in urinary excretion of CTX-I (alpha-CrossLaps), which indicates increased bone resorption in women with T2DM and osteoporosis. This marker is a product of the degradation of the newly synthesised collagen type I [15]. It has been previously reported women that the blood level of beta-CrossLaps (an isomerisation product of alpha-CrossLaps in the process of bone ageing) in postmenopausal is associated with a reduced BMD [16] and that it correlates with indicators of bone resorption in cancellous iliac bone [17].

In this study, we found a weak relationships between blood osteoprotegerin levels, excretion of CTX-I and BMD in women with T2DM. The inclusion of these markers in a multivariate discriminant analysis model does not improve the quality of the recognition of osteoporosis. The most informative predictors of osteoporosis in multivariate analysis are clinical signs: age, BMI and duration of menopause. Thus, the studied markers of bone remodeling may not be reliable indicators of osteoporosis in postmenopausal women with type 2 diabetes. However, these markers could be used for assessment the mechanisms of bone structure impairment in diabetes.

It should be noted that the highest levels of HbA1c in our study was observed in women with a normal BMD. In vitro experiments have demonstrated that high glucose concentrations increase mineralisation process in human osteoblastic cells and diminish the quality of mineralisation [14]. According to the Rotterdam study, in patients with T2DM with inadequate glycaemic control (HbA1c level ≥ 7.5%), the BMD is increased by 1.1–5.6% and fracture risk is increased by 47–63% compared to patients with HbA1c levels of <7.5% and people without T2DM [2]. Hence, a normal BMD in patients with a poor control of T2DM may create an illusion of bone health.

In recent years, the role of osteoclast and osteoblast secretion products in complex endocrine interactions between bone, adipose tissue and the pancreatic islets has been discussed. In particular, it has been shown that osteocalcin stimulates insulin secretion by β-cells and increases the insulin sensitivity of adipose tissue [10]. According to population-based studies [11, 18], low osteocalcin levels are associated with insulin resistance, metabolic syndrome and the risk of T2DM. In this study we did not find the links between the levels of osteocalcin, HbA1c and lipid metabolism parameters. It is possible that these relationships were offset by the effects of antidiabetic treatment. At the same time, the level of osteocalcin had a tendency to increase with a decrease in renal filtration function. It is known that osteocalcin is partially catabolised in the kidneys. An increase in the level of circulating osteocalcin in patients with CKD may indicate a decrease in the rate of glomerular filtration of this protein and an increase in the bone remodelling rate [19].

We recorded significantly higher blood levels of PTH in women with T2DM than in the control group and a weak positive link between PTH and HbA1c levels. It has been shown that patients with T2DM with high HbA1c levels have lower blood levels of ionised calcium and a higher secretion of parathormone in response to hypocalcaemia than patients with well-controlled T2DM [20].

Our data that women with T2DM and osteoporosis have lower fat mass and lean mass than patients with normal BMD confirm the results of earlier studies [21, 22]. However, we did not find any correlation between the concentrations of bone remodelling markers and body composition parameters.

The limitation of our study is cross-sectional design that does not revealed the causality. A number of relationships between bone remodelling markers concentrations and metabolic parameters could not be identified due to the heterogeneity of the included patients in terms of the level of glycaemia and treatment modality. We did not evaluate vitamin D levels in observed patients.

At the same time, in this study the changes in the concentrations of classical (osteocalcin, osteoprotegerin) and nonclassical (alpha-CrossLaps, CICP) markers of bone remodelling were assessed for the first time in relation to BMD, body composition and parameters of carbohydrate and lipid metabolism in postmenopausal women with T2DM.

The results of this study suggest that the development of osteoporosis in postmenopausal women with T2DM could be the consequence of both increased bone resorption and impaired bone formation.

Conclusion

Postmenopausal women with T2DM have decreased serum levels of the bone formation marker osteocalcin and the bone resorption inhibitor osteoprotegerin, and increased urinary excretion of the bone resorption marker CTX-I. In these women osteoporosis is associated with a decrease in the level of circulating osteoprotegerin and increase in the urinary excretion of CTX-I. The data give no support to hypothesis of interrelation between bone remodeling markers, metabolic parameters and TBC in T2DM.

Additional information

Funding

The study was funded by the Scientific Institute of Clinical and Experimental Lymphology.

Conflict of interest

The authors declare the absence of explicit and potential conflicts of interest associated with the publication of this article.

Author Contributions

Klimontov V.V.: Study concept and design, statistical analysis of the data, writing the text; Fazullina O.N.: Collection of clinical material, study of BMD and body composition, statistical analysis of the data, writing the text; Lykov A.P.: Laboratory tests of bone remodelling markers; Konenkov V.I.: Study concept and design, data analysis.

Acknowledgements

The authors express their gratitude to V.V. Romanov, MD, PhD, the Head of the INVITRO-Siberia Ltd. Laboratory, for assistance with urinary CTX-I measurements.

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Цитировать:

Климонтов В.В., Фазуллина О.Н., Лыков А.П., Коненков В.И. Взаимосвязь маркеров ремоделирования костной ткани с минеральной плотностью костей у женщин с сахарным диабетом 2 типа, находящихся в постменопаузе // Сахарный диабет. – 2016. – Т.19. – №5. – С. 375-382. doi: 10.14341/DM8008

To cite this article:

Klimontov VV, Fazullina ON, Lykov AP, Konenkov VI. The relationships between bone turnover markers and bone mineral density in postmenopausal type 2 diabetic women. *Diabetes mellitus*. 2016;19(5):375-382. doi: 10.14341/DM8008