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Original article

Chronic obstructive pulmonary disease severity in middle-aged and older men with osteoporosis associates with decreased bone formation

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ABSTRACT

Objectives: Chronic obstructive pulmonary disease (COPD) is a risk factor for osteoporosis. Nevertheless, much remains unclear regarding the bone metabolism dynamics associated with COPD. The present study focuses on the associations between the COPD severity and serum bone metabolism biomarkers.
Methods: We enrolled 40 patients who visited the orthopedics departments at our institutions and underwent dual-energy X-ray absorptiometry between September 2015 and December 2017. Only male osteoporosis patients over 45 years of age were included, and 5 patients were excluded due to disease or use of internal medicines affecting bone metabolism. All subjects underwent lung function testing, spine radiography, and blood tests. We measured percent forced expiratory volume in 1 second (%FEV_1), which reflects COPD severity, and we examined the relationships between %FEV_1 and serum levels of bone metabolism biomarkers.
Results: All subjects were diagnosed with osteoporosis based on T-scores. %FEV_1 correlated with body weight, body mass index (BMI), and Z-score/T-scores. %FEV_1 moderately correlated with serum levels of alkaline phosphatase (ALP), procollagen type 1 N-terminal propeptide (P1NP), and tartrate-resistant acid phosphatase 5b in the partial correlation analysis adjusted for BMI or T-score in the lumbar vertebrae. We performed a hierarchical multiple regression analysis to identify that serum ALP and P1NP were the independent explanatory variables to %FEV_1 independent of other factors.
Conclusions: The data suggest that the COPD severity in middle-aged and older men with osteoporosis associates with decreased bone formation. COPD patients may exhibit bone metabolism dynamics characterized by low bone turnover with osteogenesis dysfunction as COPD becomes severe.

1. Introduction

Many patients with chronic obstructive pulmonary disease (COPD) have osteoporosis as an extrapulmonary complication. COPD is a risk factor for osteoporosis [1,2] and patients with COPD have high prevalence rates of osteoporosis and high incidence rates of vertebral fractures [3,4]. COPD has also recently attracted the attention of researchers as a cause of secondary osteoporosis; it is considered to be the primary disease that causes secondary osteoporosis in men [5], followed by steroid use and hypogonadism.

Emphysema is the primary type of COPD found in patients, and emphysema severity negatively correlates with thoracic vertebra

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bone density [6]. Patients with severe emphysema have reduced percent forced expiratory volume in 1 second (%FEV_1) and exhibit obstructive ventilatory impairment. In general, COPD severity is determined using the GOLD classification (Global Initiative for Chronic Obstructive Lung Disease) based on %FEV_1. Bone mineral density (BMD) at various sites correlates with %FEV_1 [7].

While associations with reduced BMD in the COPD severity have been demonstrated, opinions are divided regarding bone metabolism dynamics. There are reports showing high turnover with increased bone resorption [8] and low turnover with reduced osteogenesis [9], and much remains unclear about the bone metabolism dynamics and pathophysiology of osteoporosis associated with COPD. In addition, the problem in investigating the bone metabolism dynamics of COPD-related osteoporosis is that there are various factors that affect bone metabolism, including age [10], body weight [10], smoking [11], COPD exacerbation [12], use of steroids [5], and osteoporosis treatment.

Bone metabolism markers provide important information for understanding bone metabolism dynamics. We hypothesize that COPD severity would be associated with impaired bone formation, based on our data with animal models in a previous study [13]. There is little evidence in the relationship between the COPD severity and bone metabolism markers considering the above-mentioned factors, such as age and weight. Therefore, the objective of this study is to investigate the associations between %FEV_1 and bone metabolism markers considering various factors affecting bone metabolism.

2. Methods

2.1. Subjects

This study was approved by the Ethics Review Committee for Clinical Research of our institution (approval number: H26-223). Study participants signed appropriate written consent forms after explanation and discussion of the content. Forty patients who visited the orthopedics departments at our institutions and who underwent dual-energy X-ray absorptiometry (DXA) between September 2015 and December 2017 were enrolled. We included only male patients with osteoporosis over 45 years of age. The patients were asked the presence or absence of COPD by interview with a doctor and divided into the COPD group and the control group at the time of enrollment. Subjects were randomly selected from orthopedic patients (osteoarthritis of hip or knee, lumbar canal stenosis, chronic back pain) between 2015 and 2017. Additionally, 5 of the 40 patients were excluded because they had rheumatoid arthritis ($n = 1$), bronchial asthma ($n = 2$), and use of internal medicines affecting bone metabolism, such as bisphosphonate or steroids ($n = 2$). No patients had complications with malignant tumors, severe renal/liver dysfunction or endocrine diseases, and past history of fractures within 6 months. All patients underwent lung function tests, spine radiography, and blood tests.

2.2. Lung function test

Lung function parameters were assessed using FUDAC-7C (FUKUDA DENSHI Co., Ltd., Tokyo, Japan); forced expiratory volume in 1 second (FEV_1) and forced vital capacity (FVC) were measured, and the ratio of FEV_1 to FVC (FEV_1/FVC) was calculated. All patients with COPD had $FEV_1/FVC < 70\%$. According to the GOLD guidelines, patients with %$FEV_1 > 80\%$ of their predicted maximally attained value were classified as GOLD I, patients with %FEV_1 between 50% and 80% of their predicted maximally attained value were classified as GOLD II, patients with %FEV_1 between 30% and 50% of their predicted maximally attained value were classified as GOLD III, and patients with %$FEV_1 < 30\%$ of their predicted maximally attained value were classified as GOLD IV [14].

2.3. Radiographic assessment

Radiography of the thoracic and lumbar spine was performed using RADspeed Pro (SHIMADZU CORPORATION, Kyoto, Japan), AeroDR1717 (KONICA MINOLTA, Inc., Tokyo, Japan), and CS-7 (KONICA MINOLTA, Inc., Tokyo, Japan). Vertebral bodies of T4 to L4 were independently measured by 2 investigators. Vertebral fractures were defined according to Genant et al [15]. Fractures were classified as having mild, moderate, and severe deformities. We counted the number of vertebral fractures that had a deformity classification of higher than mild.

2.4. Bone mineral density

BMDs of the femoral neck and greater trochanter of the hip and of the lumbar spine (L1–4) were measured using a HORIZON A (TOYO MEDIC Co., Ltd., Hologic Japan, Inc, Tokyo, Japan). Osteoporosis diagnosis was based on the lowest T-score of the 3 measured locations and was defined according to the World Health Organization [16]. In case of significant osteoarthritis of the lumbar spine, BMD at the lumbar spine was not assessed.

2.5. Blood sample test

The venous blood samples of all patients were obtained and measured for the following: calcium (enzymatic method), phosphorus (enzymatic method), alkaline phosphatase (ALP, JSCC reference method), total procollagen I intact N-terminal propeptide (P1NP, CLIA, SRL, Inc., Tokyo, Japan), tartrate-resistant acid phosphatase-5b (TRACP-5b, EIA, SRL, Inc.), undercarboxylated osteocalcin (ucOC, CLIA, SRL, Inc., Tokyo, Japan), 25-hydroxyvitamin D (25OHD, CLIA, SRL, Inc., Tokyo, Japan), matrix metalloproteinase 9 (MMP9) (MMP-9, Human, ELISA Kit, R&D Systems, Minneapolis, Minnesota, USA), receptor activator of NF-kappa-B ligand (RANKL) (RANK Ligand, FREE Soluble, Human, ELISA Kit, BIOMEDICA, Vienna, Austria), osteoprotegerin (OPG) (ELISA Kit, BIOMEDICA), sclerostin (Sclerostin, Human, ELISA Kit, BIOMEDICA, Vienna, Austria), dickkopf-1 (DKK1) (DKK-1, Human, ELISA Kit, BIOMEDICA), and fibroblast growth factor 23 (FGF23) (FGF23 (C-terminal and intact) Multi-Matrix ELISA Kit, BIOMEDICA). All samples were measured 3 times within the same assay lot.

2.6. Statistical analysis

Results for continuous variables are presented as mean ± standard deviation. Intergroup comparisons were performed using the Mann–Whitney *U* test or the chi-square test. The Shapiro–Wilk test was used to determine whether the variable had a standard normal distribution, and between-variable associations were analyzed using the Pearson correlation coefficient or Spearman's rank correlation coefficient. After simple correlation analysis, we performed the partial correlation analysis to adjust the effects of age, BMI, smoking status, and L1-4 T-score. Based on the results of univariate analysis, we performed a partial correlation analysis to identify the association with %FEV_1, entering the serum biomarkers that yielded a P-value of < 0.10 in the univariate analysis as an explanatory variable. After the partial correlation analysis, we performed a hierarchical multiple regression analysis. The hierarchical multiple regression analysis investigates the influence of independent variables on dependent variables by inputting the independent variable step by step. We examined the association between ALP or P1NP, which were bone-formation related markers,

and %FEV_1. P < 0.05 was considered statistically significant. We performed a power analysis and found that a minimum of 30 cases were required to perform simple correlation analysis ($\alpha = 0.05$, power = 0.8, effect size = 0.5), and a minimum of 25 cases were required to perform multiple regression analysis ($\alpha = 0.05$, power = 0.8, effect size = 0.26). All statistical analyses were performed using STATA/IC 16 (StataCorp, College Station, TX, USA).

3. Results

3.1. Demographic, lung function, T-score and serum marker data in both groups

Demographic data including, age, height, weight, BMI, and smoking status, in both groups are summarized in Table 1. All subjects were diagnosed with osteoporosis based on the value of T-score at each site. The mean BMI in the COPD group tended to be lower than that of the control group (P = 0.093). The mean values of the percent vital capacity (%VC), FEV_1/FVC and %FEV_1 in the COPD group were significantly lower than those in the control group. There were 9 cases of latent obstructive disorders in the control group. The 16 subjects in the control group were classified by the GOLD classification as follows; stage 0, 7 patients; stage I, 9 patients. The 19 subjects in the COPD group were classified by the GOLD classification as follows; stage II, 10 patients; stage III, 7 patients; stage IV, 2 patients. The vertebral-fracture number, Z-scores and T-scores between COPD and control groups also are summarized in Table 1. The number of the thoracic- or lumbar-vertebral fractures in the COPD group were significantly higher than that in the control group. The mean value of the Z-score and T-score at L1-4, total hip and femoral neck in the COPD group was significantly lower than that in the control group (Table 1). Levels of serum bone metabolism biomarkers are summarized in Table 2. Serum levels of ALP, P1NP, TRACP-5b, and sclerostin in the COPD group tended to be lower than those of the control group (P = 0.057, 0.055, 0.047, and 0.049, respectively). There were no significant differences between groups with respect to the other biomarkers.

Table 1
The characteristics of middle-aged and older male subjects enrolled in this study.

Variable	COPD (n = 19)	Control (n = 16)	P-value *
Age, yr	73.1 ± 10.8	77.4 ± 7.5	0.196
Height, cm	162.1 ± 6.3	163.6 ± 6.9	0.676
Weight, kg	56.7 ± 13.1	64.2 ± 12.8	0.123
Body mass index, kg/m^2	21.5 ± 4.7	23.8 ± 3.5	0.093
Smoking status			
Current, n (%)	5 (26.3)	4 (25.0)	0.929
Pack-year	39.4 ± 35.2	26.5 ± 27.1	0.256
Lung function			
%VC, % of predicted	75.7 ± 17.1	108.9 ± 14.8	< 0.001
FEV_1/FVC, %	55.8 ± 12.2	69.0 ± 9.4	< 0.001
%FEV_1, % of predicted	51.7 ± 16.3	95.3 ± 12.3	<0.001
Spinal X-ray			
Number of VF	1.2 ± 1.0	0.1 ± 0.3	< 0.001
Thoracic VF, n (%)	9 (47.4)	1 (6.3)	0.009
Lumbar VF, n (%)	7 (36.8)	1 (6.3)	0.032
Total VF, n (%)	13 (68.4)	2 (12.5)	0.001
DXA			
L1-4 Z-score	0.34 ± 1.1	1.93 ± 1.8	0.003
L1-4 T-score	−1.10 ± 0.9	0.54 ± 1.8	0.002
Total hip Z-score	−0.52 ± 0.7	0.84 ± 1.0	< 0.001
Total hip T-score	−1.77 ± 0.8	−0.65 ± 1.0	0.001
Femoral neck Z-score	−0.98 ± 1.0	0.32 ± 1.0	0.001
Femoral neck T-score	−2.22 ± 0.9	−1.23 ± 0.9	0.009

Values are expressed as mean ± SD unless otherwise indicated.
Comparisons were performed using the Mann-Whitney *U* test, except for qualitative variables, which were compared using the chi-squared test.
VC, vital capacity; FVC, forced vital capacity; FEV_1, forced expiratory volume in 1 s; VF, vertebral fracture; DXA, dual-energy X-ray absorptiometry.

Table 2
The serum levels of each bone-metabolism-related marker.

Variable	COPD (n = 19)	Control (n = 16)	P-value[a]
Serum markers			
Calcium, mg/dL	9.4 ± 0.4	9.3 ± 0.4	0.237
Phosphorus, mg/dL	3.3 ± 0.5	3.1 ± 0.5	0.538
ALP, U/L	202 ± 70	256 ± 64	0.057
P1NP, μg/L	49.3 ± 26.3	34.5 ± 13.1	0.055
TRACP-5b, mU/dL	293 ± 113	452 ± 234	0.047
ucOC, ng/mL	4.8 ± 3.9	7.0 ± 6.2	0.187
25OHD, ng/mL	14.5 ± 5.6	14.0 ± 4.3	0.865
MMP9, ng/mL	324 ± 113	324 ± 182	0.929
RANKL, pmol/L	0.06 ± 0.04	0.06 ± 0.05	0.547
OPG, pmol/L	6.1 ± 2.4	6.5 ± 1.9	0.722
Sclerostin, pmol/L	45.2 ± 27.5	60.4 ± 23.4	0.049
DKK1, pmol/L	43.2 ± 8.9	38.6 ± 12.6	0.420
FGF23, pmol/L	2.86 ± 4.4	1.80 ± 1.8	0.875

Values are expressed as mean ± SD unless otherwise indicated.
[a] Comparisons were performed using the Mann-Whitney *U* test. ALP, Alkaline phosphatase; P1NP, procollagen I intact N-terminal propeptide; TRACP-5b, tartrate-resistant acid phosphatase-5b; ucOC, undercarboxylated osteocalcin; 25OHD, 25-hydroxyvitamin D; MMP9, matrix metalloproteinase 9; RANKL, receptor activator of NF-kappa-B ligand; OPG, osteoprotegerin; DKK1, dickkopf-1; FGF23, fibroblast growth factor 23.

3.2. The simple correlation analysis evaluating the association with %FEV_1

%FEV_1 correlated with weight, BMI and number of vertebral fractures as well as with Z-score and T-score at each site (Table 3).

Table 3
The relationships between %FEV_1 and each parameter in middle-aged and older males.

Variable	Coefficient	P-value[a]
Age	0.206	0.236
Height	0.131	0.454
Weight	0.428	0.010
Body mass index	0.445	0.008
Smoking status		
Pack-year	−0.210	0.225
Spinal X-ray		
Number of VF	−0.693	< 0.001
DXA		
L1-4 Z-score	0.539	0.001
L1-4 T-score	0.512	0.002
Total hip Z-score	0.634	< 0.001
Total hip T-score	0.572	< 0.001
Femoral neck Z-score	0.549	< 0.001
Femoral neck T-score	0.504	0.003
Serum markers		
Calcium	−0.119	0.498
Phosphorus	−0210	0.225
ALP	0.515	0.002
P1NP	0.316	0.069
TRACP-5b	0.324	0.066
ucOC	0.280	0.114
25OHD	0.189	0.424
MMP9	−0.046	0.801
RANKL	−0.041	0.821
OPG	0.066	0.722
Sclerostin	0.322	0.072
DKK1	−0.204	0.262
FGF23	0.009	0.960

[a] Relationships were assessed by Pearson's or Spearman's rank correlation test.
VF, vertebral fracture; DXA, dual-energy X-ray absorptiometry; ALP, Alkaline phosphatase; P1NP, procollagen type 1 N-terminal propeptide; TRACP-5b, tartrate-resistant acid phosphatase-5b; ucOC, undercarboxylated osteocalcin; 25OHD, 25-hydroxyvitamin D; MMP9, matrix metalloproteinase 9; RANKL, receptor activator of NF-kappa-B ligand; OPG, osteoprotegerin; DKK1, dickkopf-1; FGF23, fibroblast growth factor 23.

Among serum markers, $\%FEV_1$ correlated with ALP. Serum levels of P1NP, TRACP-5b, and sclerostin tended to correlate with $\%FEV_1$ ($P = 0.069$, 0.066, and 0.074, respectively) (Table 3).

3.3. The age-, BMI-, pack-year- or T-score-adjusted partial correlation analysis evaluating the relationships between $\%FEV_1$ and each serum marker

We performed partial correlation analysis to adjust for the effects of age, BMI, cigarette pack-year and L1-L4 T-scores. Based on the results of univariate analysis, we entered the serum biomarkers that yielded P-values of < 0.10 in the univariate analysis as an explanatory variable. In the age-, BMI-, pack-year- or T-score-adjusted partial correlation analysis, $\%FEV_1$ correlated with the serum ALP, P1NP, and TRACP-5b. On the other hand, serum levels of sclerostin did not correlate with $\%FEV_1$ when adjusted for age, BMI, pack-year or T-score (Table 4).

3.4. Serum levels of ALP and P1NP associate with $\%FEV_1$

We performed hierarchical multiple regression analysis to determine the influence of the independent variable on the $\%FEV_1$ by inputting the independent variable step by step. The regression analysis was performed in 3 steps, and we determined whether the additional independent variables significantly increased the ΔR^2. We evaluated the influence of serum ALP or P1NP, both of which are bone-formation related markers, on $\%FEV_1$. Based on the results of simple correlation and partial correlation analysis, we determined the independent variables of each step as follows: model 1: age and BMI; model 2: age, BMI and T-score; and model 3: age, BMI, T-score, and ALP or P1NP. As a supplement, we have excluded the number of vertebral fractures from explanatory variables due to collinearity problems in the hierarchical multiple regression analysis. If the analysis was included ALP in model 3, adjusted R^2 at model 3 was 0.554 ($\Delta R^2 = 0.165$, $P = 0.003$) for the $\%FEV_1$ (Table 5A). If the analysis included P1NP in model 3, adjusted R^2 at model 3 was 0.497 ($\Delta R^2 = 0.108$, $P = 0.019$) for the $\%FEV_1$ (Table 5B). Serum levels of ALP and P1NP, which were the additional independent variable in model 3 of hierarchical multiple regression analysis with $\%FEV_1$ as dependent variables, significantly increased the ΔR^2. We found that serum ALP and P1NP were independent factors contributing to $\%FEV_1$.

Table 5
The hierarchical multiple regression analysis to evaluate the relation of (A) ALP or (B) P1NP to $\%FEV_1$.

(A)

Variable	Model 1		Model 2		Model 3	
	β	SE	β	SE	β	SE
Age	0.638	0.434	0.521	0.402	0.398	0.352
BMI	2.890**	0.973	1.645	1.020	0.753	0.927
L1-4 T-score			6.888 *	2.694	7.282**	2.345
ALP					0.160**	0.049
R^2	0.256		0.389		0.554	
ΔR^2	0		0.133 *		0.165**	

(B)

	Model 1		Model 2		Model 3	
	β	SE	β	SE	β	SE
Age	0.638	0.434	0.521	0.402	0.376	0.376
BMI	2.890**	0.973	1.645	1.020	1.490	0.944
L1-4 T-score			6.888*	2.694	6.234*	2.501
P1NP					0.420*	0.169
R^2	0.256		0.389		0.497	
ΔR^2	0		0.133*		0.108*	

Model 1: Age and body mass index (BMI), Model 2: Age, BMI and T-score at L1-4, Model 3: Age, BMI, T-score at L1-4 and serum alkaline phosphatase (ALP) or procollagen type 1 N-terminal propeptide (P1NP), *P < 0.05, **P < 0.01.

4. Discussion

We investigated the associations between $\%FEV_1$, which reflects COPD severity, and bone metabolism markers by adjusting for various factors. To the best of our knowledge, no reports have evaluated the associations between COPD severity and bone formation markers with adjustment of age, BMI, smoking status, and BMD. The primary findings of this study are as follows. First, serum ALP and P1NP, which are bone formation markers, tended to correlate with $\%FEV_1$ in the simple correlation analysis, and the relationship did not also change after adjustment for age, BMI, pack-year or T-score. Second, hierarchical multiple regression analysis revealed that serum ALP and P1NP correlated strongly to $\%FEV_1$.

Because the predicted value of FEV_1 is calculated from gender, height and age, $\%FEV_1$ is already adjusted by these factors. In fact, $\%FEV_1$ did not correlate with height or age, whereas it positively correlated with weight or BMI. Weight loss in COPD patients is associated with excessive energy expenditure by respiratory muscles, systemic inflammation, nutrition disability, and physical inactivity. Because COPD is a risk factor for sarcopenia and previous reports have shown severe muscle loss in severe COPD patients [7], some weight loss may reflect muscle loss. COPD is also a risk factor for osteoporosis and is associated with high prevalence rates of osteoporosis and incidence rates of vertebral fractures [3,4]. COPD severity correlates with BMD at various sites (radius, lumbar vertebrae, proximal femur, and femoral neck) [7]. In the present study, model 2 of the hierarchical multiple regression analysis revealed that L1-4 T-score was an explanatory factor for $\%FEV_1$ independent of BMI and age, and COPD severity strongly correlated with BMD (Tables 5A and 5B). The present study's results regarding relationship between $\%FEV_1$ and weight/BMI/T-score were not inconsistent with those of previous reports.

Some findings regarding the bone metabolism dynamics of COPD patients disagree with our results [8] while others support them [9], indicating a lack of consensus. Possible reasons for this

Table 4
The age-, BMI-, pack-year- or T-score-adjusted partial correlation analysis regarding the relationships between $\%FEV_1$ and each bone-metabolism-related marker.

Variable	Age		BMI		Pack-year		T-score	
	Coefficient	P-value[a]	Coefficient	P-value[a]	Coefficient	P-value[a]	Coefficient	P-value[a]
ALP	0.505	0.002	0.464	0.006	0.482	0.004	0.562	< 0.001
P1NP	0.431	0.012	0.445	0.009	0.437	0.011	0.437	0.011
TRACP-5b	0.399	0.024	0.512	0.003	0.430	0.014	0.422	0.016
Sclerostin	0.234	0.197	0.218	0.238	0.230	0.214	0.081	0.669

[a] Based on the results of univariate analysis, we performed a partial correlation analysis to identify independent contributing factors of $\%FEV_1$, entering the independent variables in serum biomarkers that yielded a P-value of < 0.10 in the univariate analysis. BMI, body mass index; T-score, T-score at L1-4; ALP, Alkaline phosphatase; P1NP, procollagen type 1 N-terminal propeptide; TRACP-5b, tartrate-resistant acid phosphatase-5b.

lack of consensus are that various studies examined differing subject populations, and the contributions of factors such as gender, age, and COPD exacerbations, may have affected the results. Despite the fact that Xiaomei et al [9] found that levels of markers of osteogenesis were significantly lower among elderly male COPD patients, they did not investigate the relationship between COPD severity and bone metabolism markers considering various factors. The present study, which examined middle-aged and older men, found that %FEV_1 tended to positively correlate with serum levels of P1NP and TRACP-5b, and the relationship did not change after adjustment for age, BMI, smoking status or BMD. Hierarchical multiple regression analysis revealed that serum P1NP significantly correlated with %FEV_1. These findings suggest that middle-aged and elderly male COPD patients exhibit bone metabolism dynamics involving low bone turnover with osteogenesis dysfunction as COPD becomes more severe.

We evaluated many bone metabolism biomarkers, though %FEV_1, which reflects COPD severity, only correlated with ALP, P1NP and TRACP-5b. ALP is widely distributed in organs such as the liver, bone, and small intestine, and bone-specific ALP, in particular, is used as an osteogenesis marker. It is an enzyme that plays an important role in bone mineralization, and patients who have hypophosphatasia exhibit bone fragility [17]. On the other hand, serum P1NP is generally used as a bone formation marker, and is immediately increased following administration of parathyroid hormone or anti-sclerostin antibody [18], both which are bone formation promoters. It is commonly used in Japan because it is easy to examine and does not show diurnal variation. Because P1NP is produced from the early stage of osteoblast differentiation, it is considered to be an indicator of earlier bone formation. Therefore, when COPD becomes more severe, the impaired bone formation may occur in the early stage of osteoblast differentiation. Because there have been several reports of reduced bone mineralization in COPD animal models [13,19,20], it is possible that severe COPD patients exhibit attenuated osteoblast function and declined bone mineralization. Serum TRACP-5 is generally used in Japan as a bone resorption marker. Serum levels of TRACP-5 is also easy to examine and has no diurnal variation. This serum marker decreases following the administration of bone resorption inhibitors [21–23]. Osteoclasts, which play an important role in bone resorption, have a coupling mechanism with osteoblasts, and it is interpreted that the bone turnover has been reduced, when lower values of serum TRACP-5b are shown. Although the details of these pathophysiologies in COPD patients remain still unclear, we believe that decreased bone resorption occur following impaired bone formation by any factors.

There are several concerns that should be described as study limitations. Although we mentioned that serum ALP and P1NP were independent factors contributing to %FEV_1 in a hierarchical multiple regression analysis, we could not show significant differences of those factors in simple regression analyses and the 2 group comparisons. Our small sample size is a limitation of the present study, and further studies may benefit from a larger sample size. In addition, factors affecting bone metabolism were eliminated to the greatest extent possible (age, gender, pneumonia, rheumatoid arthritis, steroids, and bisphosphonate); nevertheless, we could not exclude all effects. For example, details about the disease duration and treatment history of COPD are also unknown, and it is possible that patients with severe COPD have formerly inhaled or oral glucocorticoids. These factors may have influenced the results of this study. Additionally, bone-specific alkaline phosphatase would have been better than ALP as a bone turnover marker; therefore, we recommend that future studies use this instead.

5. Conclusions

We examined the relationships between COPD severity and several bone metabolism biomarkers using serum from middle-aged and elderly male with osteoporosis. %FEV_1, which reflects COPD severity, associates with decreased bone formation, suggesting that the bone metabolism dynamics of middle-aged and elderly men with COPD involve low bone turnover accompanied by osteogenesis dysfunction.

CRediT author statement

Manabu Tsukamoto: Formal analysis, Investigation, Resources, Writing - original draft, Writing - review & editing. **Toshiharu Mori:** Conceptualization, Writing - review & editing, Methodology. **Eiichiro Nakamura:** Resources, Funding acquisition. **Yasuaki Okada:** Investigation, Resources. **Hokuto Fukuda:** Investigation, Resources. **Yoshiaki Yamanaka:** Resources. **Ken Sabanai:** Resources. **Ke-Yong Wang:** Formal analysis. **Takeshi Hanagiri:** Resources. **Satoshi Kuboi:** Resources. **Kazuhiro Yatera:** Supervision. **Akinori Sakai:** Supervision.

Conflicts of interest

The authors declare no competing interests.

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