Investigation of the influencing factors on the efficacy evaluation of osteoporosis drugs: a meta-analysis of randomized clinical trials

Tsuyoshi Kobayashi

DP-17402

Department of Clinical Medicine (Pharmaceutical Medicine)

Graduate School of Pharmaceutical Sciences

Kitasato University

5-9-1 Shirokane, Minato-ku, Tokyo, 108-8641, Japan

Abstract

Background

The incidence of vertebral fracture is commonly used as a primary endpoint in randomized clinical trials of pharmaceutical agents for osteoporosis. The correlation between change in bone mineral density (BMD) and incidence of new vertebral fracture has been drawing attention in regard to fracture risk evaluation and drug efficacy evaluation. We firstly examined the correlation between the incidence of vertebral fracture and baseline BMD, ethnic and regional differences, and other risk factors in the placebo group by meta-regression analysis in Research 1. Next, we investigated the impact of specified risk factors on the correlation between change in BMD and incidence of new vertebral fracture by meta-regression analysis with a view to improve the accuracy of evaluation of the correlation in Research 2.

Methods

We studied a total of 21 post-menopausal osteoporosis clinical studies involving 28,425 patients treated with placebo in Research 1, and a total of 19 post-menopausal osteoporosis clinical studies involving 62,432 patients in 46 placebo or treatment groups in Research 2. These trials were identified through MEDLINE, EMBASE, and Cochrane Central Register of Controlled Trials.

Results

In research1, a univariate meta-regression showed a significant correlation between the proportion of subjects experiencing new vertebral fracture and the proportion of Caucasian subjects (coefficient = 0.223, $p = 0.045$), and the proportion of subjects with prevalent vertebral fracture (0.161, $p < 0.001$). Baseline lumber spine BMD did not

show a significant correlation. As a result of multivariate meta-regression analysis with the factors with $p < 0.2$ by the univariate meta-regression, the proportion of subjects with prevalent vertebral fracture was identified as an influencing factor (0.139, $p = 0.001$). In research2, a multivariate meta-regression analysis showed a significant correlation between the change in lumbar spine BMD and the proportion of subjects experiencing new vertebral fracture, and a lower Akaike's information criterion was obtained when the proportion of subjects with prevalent vertebral fracture was added as an explanatory variable. Significant interaction between the proportion of subjects with prevalent vertebral fracture and the change in lumbar spine BMD was shown.

Conclusion

The result of Research 1 showed that prevalent vertebral fracture was the most important factor to predict subsequent vertebral fracture. In Research 2, the change in lumbar spine BMD, not BMD T-score at one time point, was shown to have a significant correlation with the incidence of vertebral fracture. The prediction of fracture risk by change in lumbar spine BMD was improved by the adjustment with the proportion of subjects with prevalent vertebral fracture in the study population. The degree of prevalence of vertebral fracture in the population should be considered when the association between change in lumbar spine BMD and incidence of vertebral fracture is examined. We also suggest that it would be possible to evaluate drug efficacy by fracture risk estimation model based on the change in BMD adjusted by other factors such as prevalent fracture in future osteoporosis drug development.

Table of Contents

List of Tables

List of Figures

Abbreviations

AIC	Akaike's information criterion
BMD	Bone mineral density
BMI	Body mass index
CI	Confidence interval
DXA	Dual-energy X-ray absorptiometry
EMA	European Medicines Agency
FDA	The United States Food and Drug Administration
RCTs	Randomized clinical trials
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-analysis

1. Introduction

Osteoporosis is a disease characterized by low bone mass and structural deterioration of bone tissue, leading to bone fragility and an increased risk of fractures of the hip, spine, and wrist. Since 1990's, several types of osteoporosis drugs had been developed, and most recently, Romosozumab was approved in 2019 in the United States and Japan and is under review in Europe. But, after Romosozumab, new clinical trials for osteoporosis drugs seem not to have been conducted. On the other hand, between 2005 and 2025, medical care cost for osteoporotic fracture is estimated to have increased from $17 billion to $25 billion [1]. It indicates that more effective osteoporosis drug treatment is still needed to be developed.

It is known that osteoporotic hip fracture increases all-cause mortality risk five to eight fold during the first 3 months post-fracture, and the relative increase for all-cause mortality was about two to two point five fold at ≥2 years post-fracture [2]. The previous meta-analysis of cohort studies also showed a correlation between previous fracture history and subsequent fracture [3]. And, it is reported that prevalent vertebral fracture increases the risk of hip fracture [4]. Therefore, the objective of osteoporosis treatment is to prevent osteoporotic fractures, and the United States Food and Drug Administration (FDA), European Medicines Agency (EMA) and Japanese Ministry of Health, Labour and Welfare have required evidence of fracture reduction efficacy in osteoporosis drug development [5, 6]. The most commonly used fracture end point in clinical trials is incidence of morphometric vertebral fractures because of their higher incidence relative to other skeletal sites such as hip. However, the fracture events do not occur frequently in their nature, and clinical trials which set fracture events as a

primary endpoint require large numbers of patients and long duration (e.g., 2-3 years) [7]. This has been a big burden of osteoporosis drug development and one of the causes for the decline of the number of new osteoporosis drug clinical trials. Therefore, the development of models to predict fracture outcome has been discussed in various forums, including the FDA Scientific Workshop, Osteoporosis Drug Development in 2015, and the association between change in BMD and fracture reduction was often high on the agenda.

In order to investigate the influencing factors on the primary endpoint of the incidence of vertebral fracture in osteoporosis clinical trials and the impact of specified risk factors on the prediction of fracture risk by change in lumbar spine BMD, we firstly examined the correlation between the incidence of vertebral fracture and baseline BMD, ethnic and regional differences, and other risk factors in the placebo group by meta-regression analysis in Research 1. Next, we investigated the impact of specified risk factors on the correlation between change in BMD and incidence of new vertebral fracture by meta-regression analysis with a view to improve the accuracy of evaluation of the correlation in Research 2.

2. Research 1

2-1. Background and objective

There are several risk factors for development of osteoporosis and osteoporotic fracture such as age, sex, calcium and vitamin D intake, body size. Ethnicity is also one of the well-known risk factors. The prevalence of osteoporotic fracture varies among regions or ethnic groups. A systematic review on the incidence of hip fracture reported that the

prevalence of osteoporotic fracture was higher in Europe and Western Pacific regions than in Africa and Eastern Mediterranean regions [8]. In terms of ethnic differences, while the prevalence of vertebral fracture was reported to be similar between Asian and Caucasian in women aged 80 or below, the prevalence of hip fracture was lower in Asian than in Caucasian [9]. Furthermore, a cohort study in the United States reported that the prevalence of vertebral fracture was lower in black women compared with white women, even though both groups had the same bone mineral density (BMD) value [10].

BMD T-score, which is used as diagnosis criteria of osteoporosis and inclusion criteria in clinical trials for osteoporosis, is a score calculated in relation to the reference distribution of BMD in young healthy population. However, normal distribution of BMD in young healthy population varies among ethnic groups[11]. The importance of using an appropriate reference range for BMD T-score calculation in different ethnic groups or regions has been discussed for many years from the beginning of the activity for setting the bone densitometry criteria for the diagnosis of osteoporosis to the current position development conference by international society of clinical densitometry [11, 12]. When the International Osteoporosis Foundation guideline and the World Health Organization technical report defined the international reference range, ethnic and regional differences were recognized as important factors for the accurate diagnosis of osteoporosis; however, there was no appropriate data with enough sample size for each ethnic group or region to set a reference range for each of the populations and also the ethnic difference in BMD could not completely explain the ethnic difference in fracture rate. Therefore, for the sake of simplicity of the criteria for diagnosis, the guidelines used the femoral neck BMD data on Caucasian women from the NHANES III study, a large population based study of representative samples of the US population, to set the

international reference range and standard deviation for BMD T-score calculation in the diagnosis of osteoporosis [11, 13].

To align with the guidelines, the above mentioned Caucasian reference for BMD T-score has been used as inclusion criteria in most clinical trials. In some populations for which epidemiological data with enough sample size was available, on the other hand, an independent average of BMD and standard deviation different from that in the Caucasian population was shown, and a normative reference range was established based on such data in the local guidelines for diagnosis of osteoporosis, as described in the 2013 international society for clinical densitometry position development conference on bone densitometry [12]. For instance, the Japanese population showed a lower average BMD in a large population survey, and a lower reference BMD compared to the Caucasian reference range was defined as the Japanese reference BMD in the Japanese diagnostic criteria [14].

Previous clinical trials also showed the importance of choosing an appropriate reference range for BMD T-score. For example, in the FIT 2 study, which assessed the effect of Alendronate to the incidence of vertebral fracture for 3 years, the reference range that was used as inclusion criteria was updated during the study based on the NHANES III data published during the study [15]. The updated reference range corrected over-diagnosis of subjects who had been diagnosed as osteoporosis under the initial reference range. In addition, in the recent FRAME study, in which the incidence of vertebral fracture for 1 year was assessed to evaluate the effect of Romosozumab for the prevention of vertebral fracture, the Latin American population using the Caucasian reference range for diagnosis showed a low incidence of non-vertebral fracture. This result raised a question about the use of Caucasian reference range in Latino American countries [16].

In order to investigate the impact of ethnic/regional difference in osteoporosis clinical trials on the primary endpoint of the incidence of vertebral fracture, we examined, by a meta-regression analysis, the correlation between the incidence of vertebral fracture in the placebo group, which was not influenced by osteoporotic drug interventions, and baseline BMD, ethnic and regional differences or other risk factors.

2-2. Methods

Search strategy

A systematic literature search was conducted by referencing the Preferred Reporting Items for Systematic Reviews and Meta-analysis (PRISMA) statement [17]. We conducted a systematic search on June 20, 2018 to identify articles on RCTs of pharmacologic agents for postmenopausal osteoporosis compared to placebo, which were published up to December 31, 2017, through MEDLINE, EMBASE, and Cochrane Central Register of Controlled Trials.

Eligibility criteria and study selection

The systematic literature review included all RCTs assessing efficacy and safety of pharmacologic agents for postmenopausal osteoporosis that met the following criteria: (i) comparison with placebo, (ii) availability of data on the number of vertebral fracture at 3 years, (iii) inclusion of Caucasian subjects, and (iv) publication in English. First, potentially relevant trials were identified via the search algorithm including the terms of [vertebral] AND [fracture] AND [placebo]. Second, trials that met the criteria were

retrieved after reviewing the abstract. Third, full texts of the retrieved articles were reviewed, and the trials that met the above criteria were selected.

The following trials were excluded from our review: trials performed in patients with male or glucocorticoid induced osteoporosis, trials with less than 3 years study duration, extension trials from the original trials, ad hoc and sub-group analyses from the original trials, and review articles. Trials selected for review were re-checked to ensure that different trials related to the same study had not been included. The identification of relevant abstracts, the selection of trials based on the criteria described above, and the subsequent data abstraction from the full-text articles were confirmed at each step in duplicate. Any discrepancies were resolved by consensus.

Data extraction

Data on the number of subjects experiencing new vertebral fracture in 3 years in the placebo group were extracted from each study as a response variable. Potential factors affecting the new vertebral fracture such as proportion of Caucasian subjects, participation of subjects in Asian countries, proportion of subjects with prevalent vertebral fracture, baseline lumber spine BMD T-score, age, years since menopause, body mass index (BMI), and vitamin D supplementation, were also extracted as explanatory variables. We originally tried to collect information on the proportion of Asian, Black, and Hispanic subjects. However, the proportion of subjects other than Caucasian was not available in most of the studies, and we used the information whether subjects in Asian countries participated or not as an alternate factor. If only baseline lumber spine BMD actual value was available in the article, we used Hologic data to convert the value to

T-score with Caucasian reference data provided by the manufacturer. Vitamin D supplementation data was referred to the protocol based on the description whether concomitant use of vitamin D supplement was stipulated. Data not available in the original papers were supplementarily extracted from review reports by United States Food and Drug Administration or European Medicines Agency.

To confirm the quality of the study, the extracted trials were assessed using the five-point Jadad Score, which can range from 0 to 5, with higher scores indicating higher quality [18].

Statistical analysis

The proportion and 95% confidence interval (CI) for the number of subjects experiencing new vertebral fracture in 3 years in the placebo group were calculated. A random-effect model, which accounts for heterogeneity among studies, was used to conduct a meta-regression analysis to investigate potential factors affecting the incidence of new vertebral fracture. The pre-specified factors used in the meta-regression were: proportion of Caucasian subjects, participation of subjects in Asian countries, proportion of subjects with prevalent vertebral fracture, baseline lumber spine BMD T-score, age, years since menopause, body mass index (BMI), and vitamin D supplementation.

A univariate meta-regression was first performed to identify potential factors affecting the incidence of new vertebral fracture. Factors with $p < 0.2$ in the univariate analysis were further analyzed by multivariate meta-regression. A statistically significant difference was defined as $p<0.05$ in the multivariate meta-regression. All the analyses were performed using R software version 3.4.3[19].

2-3. Results

Search results and study characteristics

One thousand, six hundred and fifty-two articles were identified during the literature search in EMBASE and MEDLINE and 587 articles were identified from the search in Cochrane Central Register of Controlled trials. Five hundred and fifty-eight duplicates were identified and eliminated. Of the remaining 1681 potentially relevant articles, 1626 were excluded after reviewing the titles and abstracts (Figure 1).

The full texts of the remaining 55 articles were reviewed, and 21 studies in 20 articles were ultimately included in the analysis. We used a total of 21 studies involving 28425 patients in the placebo group that met the selection criteria for the analyses. The main characteristics of the 21 studies are described in Table 1. Three of 21 studies restricted enrollment of ethnic populations other than Caucasian, and 5 studies restricted enrollment of subjects without prevalent vertebral fracture by the inclusion criteria. All the extracted trials had a score of 5 in the Jadad Score.

Figure 1. Flow diagram of study selection in Research 1

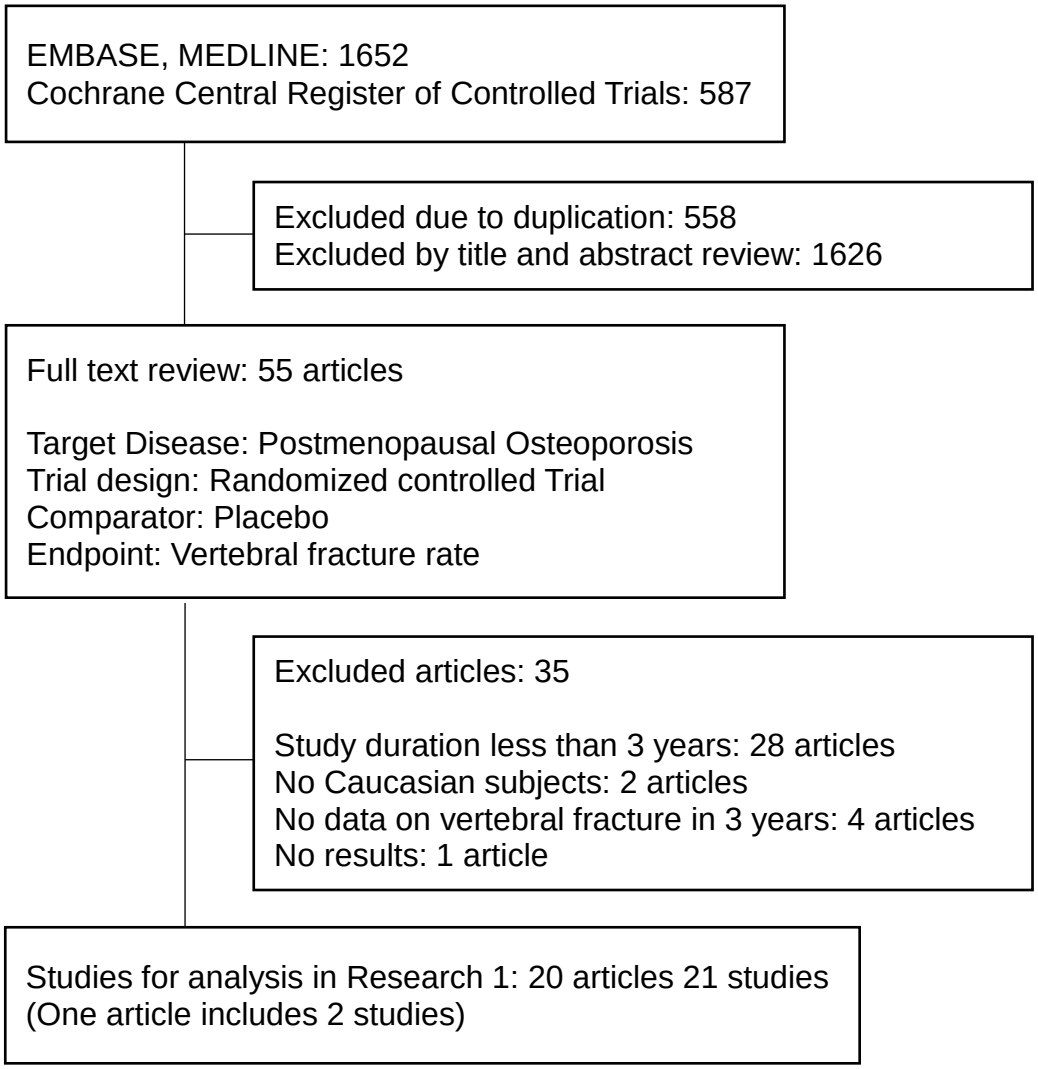


Table 1. Studies included in the analysis of Research 1

Reference	Number of subjects experiencing new vertebral fracture in 3 years	Number of subjects at baseline	Proportion of Caucasian subjects	Participation of subjects in Asian countries	Age	Baseline lumber spine BMD T-score	Proportion of subjects with prevalent vertebral fracture	BMI	Years since menopause	VD supplementation
Steven T. Harris et al. [20] 1993	32	184	0.980	N	68.5	-1.70	1.000	NA	21.3	N
Uri A. Liberman et al. [21] 1995	22	355	0.874	N	64.0	-3.06	0.212	24.1	17.0	N
Dennis M Black et al. [22] 1996	145	965	0.970	N	71.0	-2.34	1.000	25.6	NA	Y
Steven T. Harris et al. [23] 1999	93	678	0.950	N	68.0	-2.40	0.790	27.5	24.0	Y
Bruce Ettinger et al. [24] 1999	68	1522	0.957	Y	69.0	-2.54	0.101	25.8	21.0	Y
Bruce Ettinger et al. [24] 1999	163	770	0.957	Y	65.0	-2.72	0.884	25.0	18.0	Y
J. Y. Reginster et al. [25] 2000	89	346	0.990	N	71.0	-2.77	1.000	NA	25.0	Y
Charles H Chesnut III et al. [26] 2000	70	270	0.980	N	68.2	-1.79	0.790	24.7	22.0	Y
J. Y. Reginster et al. [27] 2001	114	603	0.998	N	67.4	-2.93	0.819	25.3	19.5	N
Peter Alexandersen et al. [28] 2001	11	239	1.000	N	63.4	-2.86	0.000	24.7	NA	N
Charles H Chesnut III et al. [29] 2004	73	975	0.985	N	69.0	-2.80	0.930	26..2	20.8	Y
R. Recker et al. [30] 2004	95	949	0.990	N	67.0	-2.79	1.000	25.2	20.8	Y
Pierre J. Meunier et al. [31] 2004	237	723	1.000	N	69.2	-3.60	1.000	26.2	21.6	Y
J. Y. Reginster et al. [32] 2005	367	1823	1.000	N	76.7	-3.13	0.349	NA	28.4	Y
Dennis M. Black et al. [33] 2007	310	2853	0.791	Y	73.0	-2.34	0.642	25.4	NA	Y
Stuart L Silverman et al. [34] 2008	60	1741	0.871	Y	66.5	-2.40	0.564	26.3	19.5	Y
Steven R. Cummings et al. [35] 2008	126	2257	0.990	N	68.2	-2.90	0.260	25.7	NA	Y
Steven R. Cummings et al. [36] 2009	264	3691	0.929	N	72.3	-2.84	0.234	26.0	24.2	Y
Steven R. Cummings et al. [37] 2010	176	2744	0.743	Y	67.5	-3.00	0.282	25.4	NA	Y
Steven R. Cummings et al. [38] 2011	147	2612	0.351	Y	67.6	-2.93	0.270	26.0	NA	Y
Kim Henriksen et al. [39] 2016	99	2125	0.663	Y	67.0	NA	0.214	26.0	NA	Y

Abbreviations: BMD, bone mineral density; BMI, body mass index; N, no; NA, not applicable; Y, yes.

Univariate Meta-Regression

Univariate meta-regression showed significant correlations between the "proportion of subjects experiencing new vertebral fracture in 3 years" and the "proportion of Caucasian subjects" (coefficient = 0.223, p = 0.045), and the "proportion of subjects with prevalent vertebral fracture" (0.161, p < 0.001) (Table 2). Other explanatory variables including "baseline lumber spine BMD T-score" did not show a significant correlation with the "proportion of subjects experiencing new vertebral fracture in 3 years". Even though a statistically significant correlation was not observed, the coefficient estimates for each variable were consistent with the general perception of the relation of each variable with vertebral fracture, meaning that risk factors such as "age" showed a positive correlation and preventive factors such as "BMI" showed a negative correlation.

Table 2. Univariate meta-regression analysis

	Estimate	S.E.	*p* value
Proportion of Caucasian subjects	0.223	0.112	0.045**
Participation of subjects in Asian countries	-0.068	0.037	0.065*
Proportion of subjects with prevalent vertebral fracture	0.161	0.040	<0.001**
Baseline lumber spine BMD T-score	-0.004	0.046	0.936
Age	0.008	0.006	0.169*
Years since menopause	0.009	0.009	0.303
BMI	-0.005	0.027	0.867
Vitamin D supplementation	0.011	0.049	0.817

Abbreviations: BMD, bone mineral density; BMI, body mass index; SE, standard error.

*p value < 0.2, ** p value < 0.05

Multivariate Meta-Regression

Factors that showed a correlation with the "proportion of subjects experiencing new

vertebral fracture in 3 years" with p<0.2 in the univariate meta-regression were further analyzed by multivariate meta-regression (Table 3). Because the "participation of subjects in Asian countries" seemed to have a correlation with the "proportion of Caucasian subjects", it was excluded from the multivariate analysis. As a result, the "proportion of subjects with prevalent vertebral fracture" was identified as an influencing factor (coefficient = 0.139, p = 0.001).

When we conducted the analysis using "participation of subjects in Asian countries" instead of "proportion of Caucasian subjects", the result was not changed (data not shown). We also obtained a similar result by the analysis using additional data from 3 studies for which the fracture data were not published as an article or were available only in charts in the article (data not shown).

Table 3. Multivariate meta-regression analysis

	Estimate	S.E.	*p* value
Proportion of Caucasian subjects	0.088	0.097	0.361
Proportion of subjects with prevalent vertebral fracture	0.139	0.044	0.001**
Age	0.006	0.005	0.239

Abbreviations: SE, standard error.

**p value < 0.05

2-4. Discussion

The result of the present study showed that, among the several factors investigated, "prevalent vertebral fracture" was the highest risk factor for the incidence of new vertebral fracture. The previous meta-analysis of cohort studies showed a correlation between previous fracture history and subsequent fracture [3]. Also, the diagnosis

guidelines of osteoporosis [11, 14] as well as the inclusion criteria of clinical trials of pharmaceutical agents for osteoporosis define the prevalent osteoporotic fracture including vertebral fracture as an important criterion of osteoporosis. The result of the present study is consistent with the previous report, clinical guidelines and clinical trial practice, and reconfirms the importance of the prevention of first vertebral fracture.

In univariate meta-regression analysis, while “proportion of Caucasian subjects” showed a significant positive correlation with the incidence of new vertebral fracture, “participation of subjects in Asian countries” showed a negative correlation, although it was not statistically significant. Prevalence of osteoporotic fracture was reported to be higher in Europe and Asia than in Africa or Eastern Mediterranean [8]. In addition, similar prevalence of vertebral fracture was reported between Asian and Caucasian [9]. Considering these previous reports, we could assume that the more Caucasian or Asian participants in clinical trials, the higher the incidence of new vertebral fracture. However, the present study showed different results. It is possible that the difference was led by the use of the Caucasian reference in the inclusion criteria of clinical trials, which resulted in over-diagnosis of osteoporosis in the Asian population. This is because, as the Japanese reference showed, the Asian average BMD is presumably lower than that in the Caucasian reference range. Taken together, we would suggest that the ethnic/regional difference should be considered as one of the important factors that influence the primary endpoint of osteoporosis study when the Caucasian reference range is used in the clinical trial.

In the present study, “baseline lumber spine BMD T-score” and “age” did not show a significant correlation with the incidence of vertebral fracture, despite the fact that low BMD and old age are important risk factors of osteoporotic fracture and that the BMD is

used as a diagnosis criterion. We considered that one of the reasons that “baseline lumber spine BMD T-score” did not show a significant correlation was the use of Caucasian reference range. Previous studies showed that the use of the Caucasian reference range for BMD T-score calculation in non-Caucasian populations led to unexpected outcomes [15, 16]. Another reason would be that the BMD T-score at baseline does not fully predict the risk of fracture in the future, although it is related to the risk of fracture at the timing of measuring BMD. We considered that the reason that “age” did not show a significant correlation with the incidence of vertebral fracture was that the target subjects in postmenopausal osteoporosis studies were elderly outpatients and the mean age of subjects in each study distributed too narrowly to show a significant correlation compared with the previous epidemiological data.

In order to discuss the appropriateness of using the Caucasian reference range for BMD T-score calculation in non-Caucasian populations, it might be interesting to include the results of studies without Caucasian populations. We found two non-Caucasian studies which have the fracture data at 3 years but, these two studies were in the Japanese and the Japanese reference range was used for the BMD T-score calculation. More RCTs which evaluate the incidence of vertebral fracture in non-Caucasian population using the Caucasian reference range for BMD T-score calculation would help to discuss the influence of ethnic differences in a broad sense in the future.

3. Research 2

3-1. Background and objective

The FDA and EMA have required evidence of fracture reduction efficacy in osteoporosis drug development, and have reservations about the use of BMD alone as a surrogate for fracture outcomes in confirmatory clinical trials [5, 6]. The development of models to predict fracture outcome has been discussed in various forums, including the FDA Scientific Workshop, Osteoporosis Drug Development in 2015, and the association between change in BMD and fracture reduction was often high on the agenda.

The post hoc analysis of clinical studies with Strontium Ranelate reported no association between lumbar BMD change and incidence of vertebral fracture [40]. It was also reported that when interpreting the association between the increase in BMD with fracture risk reduction by Strontium Ranelate treatment, it is necessary to consider the fact that part of the changes in BMD by Strontium Ranelate treatment was caused by the higher atomic number of strontium (Z = 38) compared with calcium (Z = 20) [41]. When BMD is measured by Dual-energy X-ray absorptiometry (DXA), strontium atoms in the bone attenuate X-rays more strongly than calcium, causing overestimation of BMD [42].

In contrast, a larger increase in lumbar spine BMD by Alendronate treatment showed a significant correlation with lower risk of vertebral fracture [43]. A systematic review which examined the association between relative risk of vertebral fracture compared with placebo and increase in BMD, also concluded that a larger increase in BMD tended to have greater anti fracture efficacy [44]. In these studies however, the influences of other factors on the association were not considered. For instance, the differences in the ratio

of subjects with prevalent fracture among studies were masked in these reports. A previous meta-analysis of 11 cohort studies, where osteoporotic fracture history and subsequent fracture for individual subjects were examined, showed a correlation between previous fracture history and subsequent fracture [3]. The diagnosis guidelines of osteoporosis [11, 14] as well as the inclusion criteria of clinical trials of drugs for osteoporosis define prevalent osteoporotic fracture, including vertebral fracture, as an important diagnostic criterion of osteoporosis. In Research 1, we examined the correlation between the incidence of vertebral fracture in the placebo group and several demographic factors at baseline by meta-regression analysis. Results showed that the proportion of subjects with prevalent vertebral fracture had a correlation with the incidence of vertebral fracture, but the baseline BMD T-score did not show a significant correlation with the incidence of vertebral fracture [45]. These results indicated that baseline BMD T-score do not predict the incidence of fracture in the 3 year study period and suggest the need to evaluate the correlation between change in lumbar spine BMD and incidence of vertebral fracture.

In Research 2, we investigated the impact of the prevalence of vertebral fracture on the correlation between the change in lumbar spine BMD and the incidence of new vertebral fracture by a meta-regression analysis, with a view to improve the evaluation of the correlation.

3-2. Methods

Search strategy

We conducted a systematic search on October 1, 2019 to identify articles on RCTs of

pharmacologic agents for postmenopausal osteoporosis compared to placebo, through MEDLINE, EMBASE, and Cochrane Central Register of Controlled Trials. This was conducted by referencing the PRISMA statement [17].

Eligibility criteria and study selection

The systematic literature review included all RCTs assessing the efficacy and safety of pharmacologic agents for postmenopausal osteoporosis that met the following criteria: (i) study compared with placebo, (ii) data on the number of vertebral fracture at 3 years available, (iii) study in which Caucasian subjects were enrolled , and (iv) report published in English. First, potentially relevant trials were identified via a search algorithm including the terms of [vertebral] AND [fracture] AND [placebo]. Next, trials that met the above-mentioned criteria were selected through the abstract and full text review.

The following trials were excluded: trials performed in patients with males, trials performed in patients with glucocorticoid induced osteoporosis, trials with less than 3 years of study duration, extension trials from original trials, ad-hoc and sub-group analyses from original trials, and review articles. Trials selected for review were re-checked to ensure that different trials related to the same study were not included. Identification of relevant abstracts, selection of trials based on the criteria described above, and subsequent data abstraction from the full-text articles were confirmed at each step in duplicate. Any discrepancies were resolved by consensus.

Data extraction

We extracted the data on the number of subjects experiencing new vertebral fracture in 3

years, in both the treatment and placebo groups from each study as a response variable. As explanatory variables, we extracted data on the percent change in lumbar spine BMD from baseline at 3 years, baseline lumbar spine BMD T-score, and proportion of subjects with prevalent vertebral fracture. We excluded the study groups treated by Strontium Ranelate, because previous reports showed that part of the changes in BMD by Strontium Ranelate treatment was caused by the higher atomic number of strontium compared with calcium [41, 42]

The data that was not available in original articles were supplementary extracted from review reports by the FDA or EMA. If only the baseline BMD actual value was available in the article, we used the Hologic data to convert the value to T-score with Caucasian reference data provided by the manufacturer.

To confirm the quality of our study, the extracted trials were assessed by all the authors using the five-point Jadad Score, which ranges from 0 to 5, with higher scores indicating higher quality [18].

Statistical analysis

The proportion and 95% CI for the number of subjects experiencing new vertebral fracture in the 3 year study period were calculated. A random-effect model, which accounts for heterogeneity among studies, was used to conduct a meta-regression analysis to investigate the correlation between percent change in lumbar spine BMD and proportion of subjects experiencing new vertebral fractures.

In order to analyze the influence of the proportion of subjects with prevalent vertebral fracture on the correlation between percent change in lumbar spine BMD and incidence

of new vertebral fractures, we performed a multivariate meta-regression analysis with or without including the proportion of subjects with prevalent vertebral fracture as a covariate, and calculated Akaike's information criterion (AIC) for each model. The factors used in the meta-regression were the percent change in lumbar spine BMD from baseline at 3 years, baseline lumbar spine BMD T-score, and proportion of subjects with prevalent vertebral fracture.

In the subgroup analysis, we divided the studies into three by tertile of the proportion of subjects with prevalent vertebral fracture and performed multivariate meta-regression analysis in the subgroups of the higher and lower tertiles without including the proportion of subjects with prevalent vertebral fracture as a covariate. We analyzed the interaction between the subgroups and the percent change in lumbar spine BMD from baseline at 3 years on the proportion of subjects experiencing new vertebral fractures.

All the analyses were performed using R software version 3.4.3 [19].

3-3. Results

Search results and study characteristics

One thousand, seven hundred and sixty-nine articles were identified through the literature search in EMBASE and MEDLINE, and 797 articles were identified from the search in Cochrane Central Register of Controlled trials. Seven hundred and eighty-three duplicates were identified and eliminated. Of the remaining 1783 potentially relevant articles, 1728 were excluded after reviewing the titles and abstracts based on the criteria (Figure 2).

The full texts of the remaining 55 articles were reviewed, and 19 studies in 18 articles

were finally included in the analysis. We used a total of 19 studies involving 62432 patients in 46 placebo or treatment groups. The characteristics of the 46 placebo or treatment groups in 19 studies are described in Table 4.

As with Strontium Ranelate, effect of Fluoride treatment on fracture and BMD were also discussed in the FDA Scientific Workshop. However, the Fluoride study was excluded due to lack of published data on the actual number of subjects experiencing new vertebral fracture in 3 years.

All the extracted trials had a score of 5 in the Jadad Score.

Figure 2 Flow diagram of study selection in Research 2

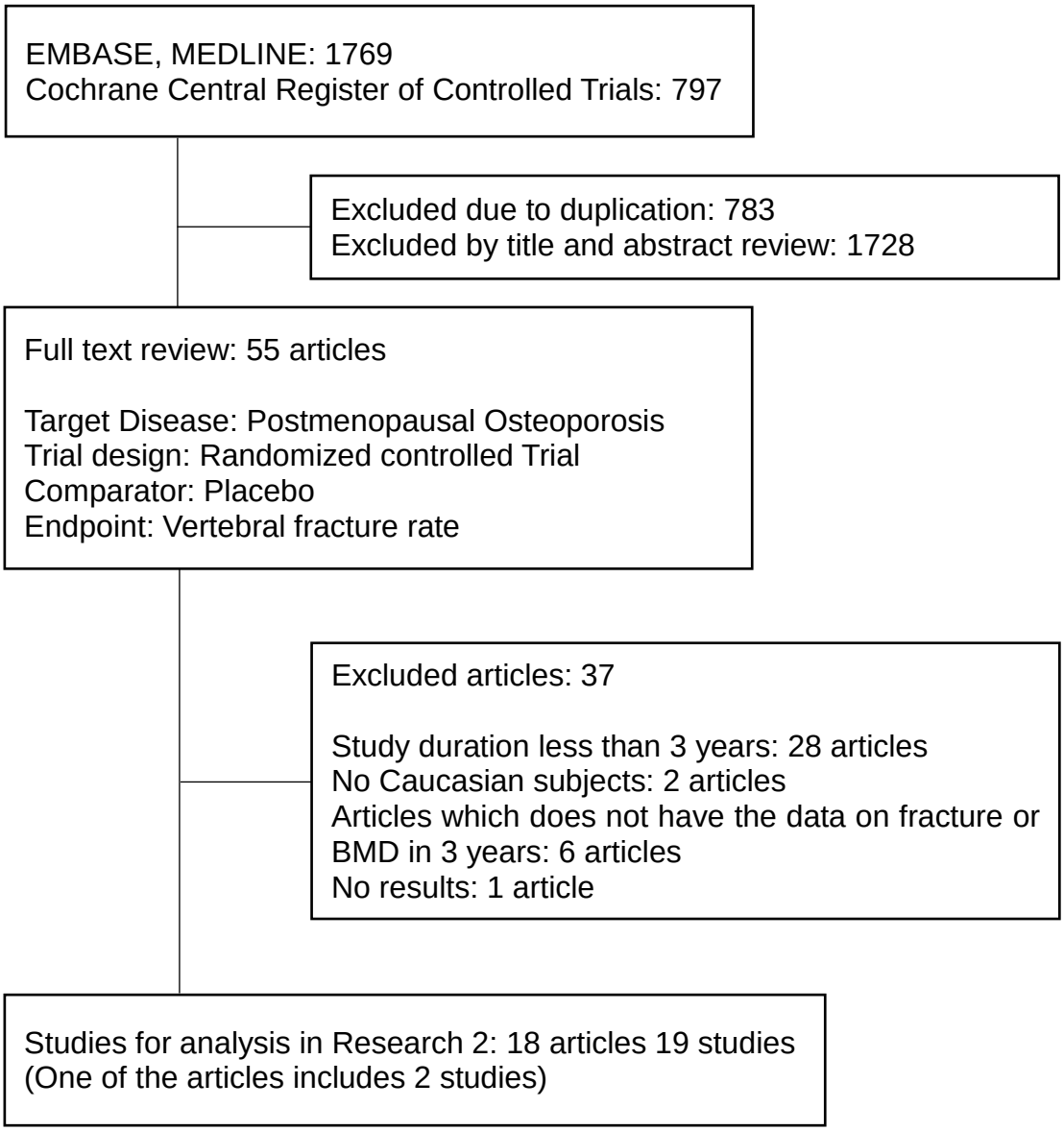


Table 4. Studies included in the analysis of Research 2

Reference	Treatment Group	Number of subjects experiencing vertebral fracture in 3 years	Number of subjects at baseline for vertebral fracture assessment	Proportion of subjects with prevalent vertebral fracture	Percent change of lumbar spine BMD from baseline at 3 years	Baseline lumbar spine BMD T-score
Peter Alexandersen et al. 2001 [28]	Placebo	11	239	0.000	0.10%	-2.86
Peter Alexandersen et al. 2001 [28]	Iprifravone	11	234	0.000	0.80%	-1.89
Bruce Ettinger et al. 1999 [24]	Placebo	68	1522	0.101	0.20%	-2.54
Bruce Ettinger et al. 1999 [24]	Raloxifene 120 mg	42	1512	0.113	2.76%	NA
Bruce Ettinger et al. 1999 [24]	Raloxifene 60 mg	35	1490	0.113	2.86%	-2.48
Uri A. Liberman et al. 1995 [21]	Placebo	22	355	0.212	-0.70% [a)]	-3.06
Uri A. Liberman et al. 1995 [21]	Alendronate 5 mg, 10 mg, or 20 mg then 5 mg	17	526	0.202	6.67% [a)]	-3.06
Kim Henriksen et al. 2016 [39]	Placebo	99	2125	0.214	0.18%	NA
Kim Henriksen et al. 2016 [39]	Oral Salmon Calcitonin	94	2064	0.253	1.02%	NA
Steven R. Cummings et al. 2009 [36]	Placebo	264	3691	0.234	0.60%	-2.84
Steven R. Cummings et al. 2009 [36]	Denosumab	86	3702	0.238	9.40%	-2.82
Steven R. Cummings et al. 2008 [35]	Placebo	126	2257	0.260	1.30% [a)]	-2.90
Steven R. Cummings et al. 2008 [35]	Tibolone	70	2249	0.270	6.50% [a)]	-2.90
Steven R. Cummings et al. 2011 [38]	Placebo	147	2612	0.270	0.90%	-2.93
Steven R. Cummings et al. 2011 [38]	Arzoxifene	209	2640	0.272	3.70%	-2.95
Steven R. Cummings et al. 2010 [37]	Placebo	176	2744	0.282	1.33%	-3.00
Steven R. Cummings et al. 2010 [37]	Lafosoxifene 0.25 mg	124	2734	0.282	4.62%	-3.00
Steven R. Cummings et al. 2010 [37]	Lafosoxifene 0.5 mg	105	2748	0.283	4.68%	-3.00
Stuart L Silverman et al. 2008 [34]	Placebo	60	1741	0.564	0.88%	-2.4
Stuart L Silverman et al. 2008 [34]	Raloxifene 60 mg	43	1849	0.563	2.96%	-2.4
Stuart L Silverman et al. 2008 [34]	Bazedoxifene 20 mg,	37	1724	0.561	2.21%	-2.4
Stuart L Silverman et al. 2008 [34]	Bazedoxifene 40 mg,	38	1686	0.559	2.38%	-2.4
Dennis M. Black et al. 2007 [33]	Placebo	310	2853	0.642	0.24%	-2.34
Dennis M. Black et al. 2007 [33]	Zoledronate	92	2822	0.623	6.95%	-2.34
Charles H Chesnut III et al. 2000 [26]	Placebo	70	270	0.790	0.50% [a)]	-1.79

Charles H Chesnut III et al. 2000 [26]	Spray Salmon Calcitonin 100 IU	59	273	0.750	1.00% [a)]	-1.88
Charles H Chesnut III et al. 2000 [26]	Spray Salmon Calcitonin 200 IU	51	287	0.790	1.00% [a)]	-1.79
Charles H Chesnut III et al. 2000 [26]	Spray Salmon Calcitonin 400 IU	61	278	0.810	1.60% [a)]	-1.88
Steven T. Harris et al. 1999 [23]	Placebo	93	678	0.790	1.10%	-2.40
Steven T. Harris et al. 1999 [23]	Risedronate 5 mg	61	696	0.800	5.40%	-2.4
Bruce Ettinger et al. 1999 [24]	Placebo	163	770	0.884	1.12%	-2.72
Bruce Ettinger et al. 1999 [24]	Raloxifene 120 mg	82	765	0.900	2.55%	NA
Bruce Ettinger et al. 1999 [24]	Raloxifene 60 mg	113	769	0.900	2.23%	-2.75
Charles H Chesnut III et al. 2004 [29]	Placebo	73	975	0.930	1.30%	-2.80
Charles H Chesnut III et al. 2004 [29]	Oral Ibandronate 2.5 mg daily	37	977	0.940	6.50%	-2.80
Charles H Chesnut III et al. 2004 [29]	Oral Ibandronate 20 mg intermittent	39	977	0.940	5.70%	-2.70
R. Recker et al. 2004 [30]	Placebo	95	949	1.000	0.91%	-2.79
R. Recker et al. 2004 [30]	Ibandronate iv 0.5 mg	76	950	0.980	3.82%	-2.79
R. Recker et al. 2004 [30]	Ibandronate iv 1.0 mg	80	960	0.980	4.73%	-2.70
Steven T. Harris et al. 1993 [20]	Placebo	32	184	1.000	1.03%	-1.70
Steven T. Harris et al. 1993 [20]	Cyclic Etidronate	28	196	1.000	5.08%	-1.52
Dennis M Black et al. 1996 [22]	Placebo	145	965	1.000	1.50% [a)]	-2.34
Dennis M Black et al. 1996 [22]	Alendronate 5 mg then 10 mg	78	981	1.000	8.00% [a)]	-2.34
J. Y. Reginster et al. 2000 [25]	Placebo	89	346	1.000	1.00% [a)]	-2.77
J. Y. Reginster et al. 2000 [25]	Risedronate 5 mg	53	344	1.000	7.00% [a)]	-2.84
Pierre J. Meunier et al. 2004 [31]	Placebo	237	723	1.000	-1.70%	-3.60
Pierre J. Meunier et al. 2004 [31]	Strontium Ranelate [b)]	150	719	1.000	12.70%	-3.50

Abbreviations: BMD, bone mineral density; IV, intravenous; NA, not applicable

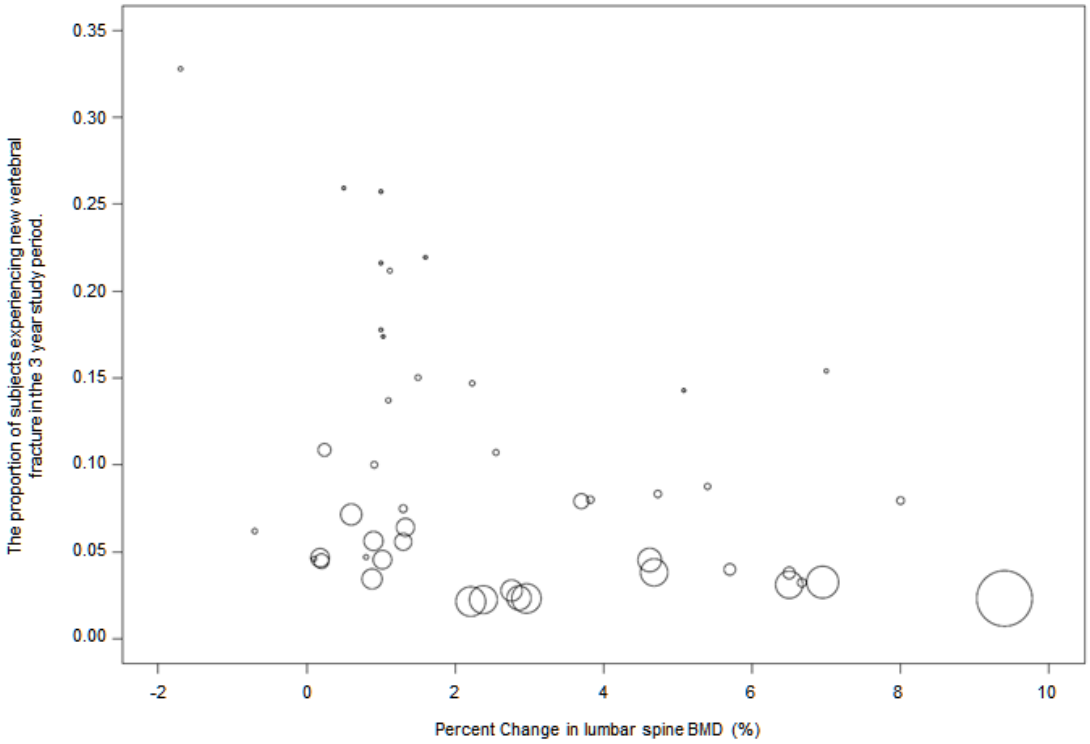
a) data estimated form bar chart or line chart in the article, b) Strontium Ranelate treated group was excluded from this analysis.

Gray masked study groups are lower and higher tertile of subgroup analysis

Multivariate meta-regression

The proportion of subjects experiencing new fracture were plotted against the percent change in lumbar spine BMD for each study (Figure 3). The multivariate meta-regression analysis showed a significant correlation between the percent change in lumbar spine BMD from baseline at 3 years, and the proportion of subjects experiencing new vertebral fracture, regardless of whether the proportion of subjects with prevalent vertebral fracture was included or not as an explanatory variable. The analysis with the proportion of subjects with prevalent vertebral fracture as an explanatory variable showed a lower AIC than the analysis without it (Table 5). When we added the Strontium Ranelate treated group in the data set, the percent change in lumbar spine BMD from baseline at 3 years did not show a significant correlation with the proportion of subjects experiencing new vertebral fractures in the model without the proportion of subjects with prevalent vertebral fracture as an explanatory variable (data not shown).

Figure 3. Scatter plot of whole study groups



Circle size reflects sample size of each study group.

Table 5. Correlation between percent change in BMD from baseline at 3 years and incidence of vertebral fracture

Analysis without the proportion of subjects with prevalent vertebral fracture as an explanatory variable			
AIC	-88.899		
Explanatory variables for multivariate meta-regression analysis	Regression coefficient	S.E.	*p* value
Percent change in BMD from baseline at 3 years	-1.127	0.406	0.006*
Lumbar spine BMD T-score at baseline	0.031	0.025	0.208
Analysis with the proportion of subjects with prevalent vertebral fracture as an explanatory variable			
AIC	-105.089		
Explanatory variables for multivariate meta-regression analysis	Regression coefficient	S.E.	*p* value
Percent change in BMD from baseline at 3 years	-1.264	0.310	< 0.001*
Lumbar spine BMD T-score at baseline	0.003	0.020	0.882
Proportion of subjects with prevalent vertebral fracture	0.133	0.025	< 0.001*

Abbreviations: BMD, bone mineral density; S.E., standard error; AIC, Akaike information criterion

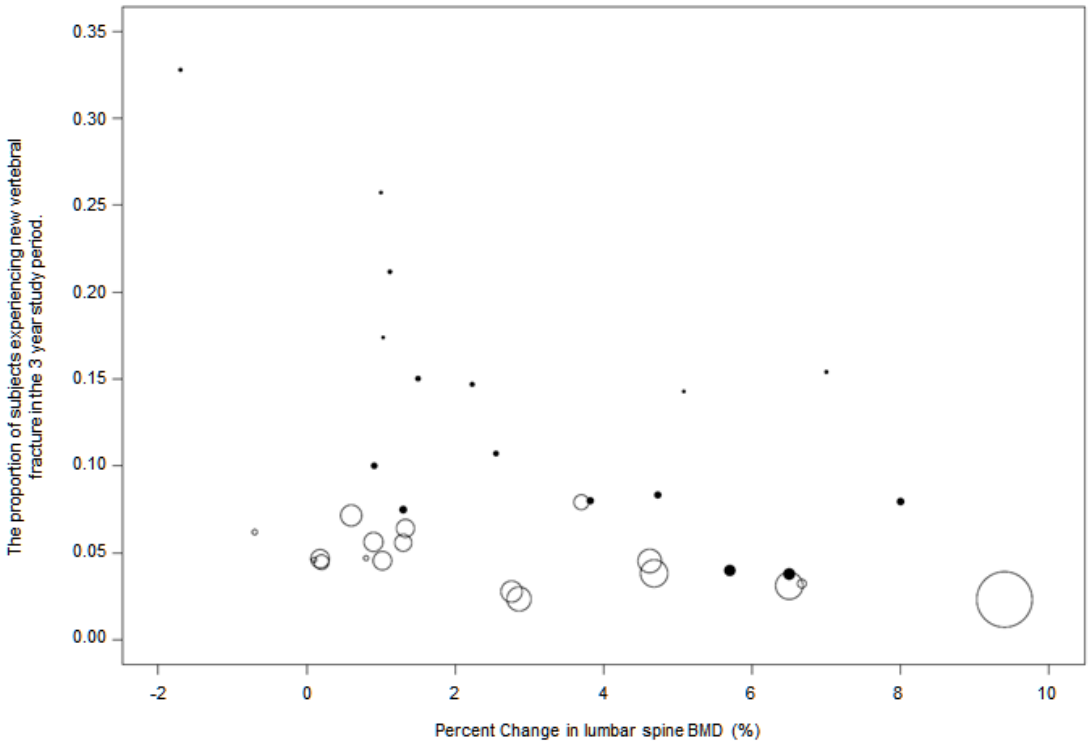
*$p<0.05$

Subgroup analysis divided by tertile of proportion of subjects with prevalent vertebral fracture

We divided the studies by tertile of the proportion of subjects with prevalent vertebral fracture (higher tertile ≥ 0.884 and lower tertile < 0.283, Table 4 and Figure 4). The percent change in lumbar spine BMD from baseline at 3 years showed a significant correlation with the proportion of subjects experiencing new vertebral fracture in both subgroups of lower and higher tertile. We found a significant interaction between the

subgroups and the percent change in lumbar spine BMD from baseline at 3 years (Table 6).

Figure 4. Scatter plot of study groups for sub-groups



Circle size reflects sample size of each study group.
Black circle: Higher tertile subgroup (the proportion of subjects with prevalent fracture ≥ 0.884)
White circle: Lower tertile subgroup (the proportion of subjects with prevalent fracture < 0.283)

Table 6. Subgroup analysis by tertile of the ratio of subjects with prevalent vertebral fracture on the correlation between percent change in BMD from baseline at 3 years and incidence of vertebral fracture

Multivariate meta-regression analysis by lower tertile	Regression coefficient	S.E.	*p* value
Percent change in BMD from baseline at 3 years	-0.423	0.119	< 0.001 *
Lumbar spine BMD T-score at baseline	-0.028	0.014	0.042 *
Multivariate meta-regression analysis by higher tertile	Regression coefficient	S.E.	*p* value
Percent change in BMD from baseline at 3 years	-1.877	0.637	0.003 *
Lumbar spine BMD T-score at baseline	-0.010	0.038	0.795
Interaction analysis	Regression coefficient	S.E.	*p* value
Percent change in BMD from baseline at 3 years	-0.376	0.390	0.334
Lumbar spine BMD T-score at baseline	-0.014	0.022	0.545
Difference of subgroups	0.137	0.024	< 0.001*
Interaction between the subgroups divided by proportion of subjects with prevalent vertebral fracture and the percent change in BMD from baseline at 3 years	-1.473	0.595	0.013 *

Abbreviations: BMD, bone mineral density; S.E., standard error

*$p<0.05$

3-4. Discussion

The results of the present study showed a significant correlation between the percent change in lumbar spine BMD from baseline at 3 years and the proportion of subjects experiencing new vertebral fractures regardless of the adjustment with the proportion of subjects with prevalent vertebral fracture. Previous ad-hoc analysis of clinical trial data of osteoporosis drugs and a systematic review reported that larger increase in BMD

tended to have a greater anti-fracture efficacy [43, 44]. The present result was consistent with these reports. We suggest that the increase in lumbar spine BMD correlates with the prevention of new vertebral fracture under the circumstances where the osteoporosis agent does not affect the DXA measurement like Strontium Ranelate.

Although the change in lumbar spine BMD showed a significant correlation with the incidence of new vertebral fracture regardless of the adjustment with the proportion of subjects with prevalent vertebral fracture, the AIC of the analysis with the adjustment was lower than that without the adjustment. This result indicated that the model with the adjustment predicts more appropriately the incidence of new vertebral fracture than the model without the adjustment. We considered several factors leading to this result. First, as reported in previous studies including the meta-analysis of cohort studies and our previous meta-regression analysis in the placebo group in clinical trials, the prevalence of vertebral fracture has a significant correlation with the incidence of subsequent vertebral fracture [3, 45]. These results indicate that the higher prevalence of vertebral fracture, the higher incidence of new vertebral fracture we observe. Therefore, difference in the prevalence of vertebral fracture among study populations should be considered when we compare the fracture prevention effect among studies. Second, the vertebral fracture itself has influence on BMD measurement. L1 is one of the sites in which fractures most frequently occur [46]. One or two fractures in the lumbar spine increase BMD [47]. The international society of clinical densitometry has recommended that anatomically abnormal vertebrae should be excluded from analysis if they are clearly abnormal and

non-assessable within the resolution of the system; or if there is more than a 1.0 T-score difference between the vertebra in question and the adjacent vertebrae [48]. It can be interpreted that vertebral fracture affects the measurement of lumbar spine BMD, and the prevalence of vertebral fracture reduces the accuracy of fracture risk prediction by BMD. In the present results of subgroup analysis by tertile of the proportion of subjects with prevalent vertebral fracture, however, the change in BMD showed a significant correlation with the incidence of new vertebral fracture in both the higher and the lower tertile group without the adjustment with the proportion of subjects with prevalent vertebral fracture. Therefore, we suggest that the main factor leading to the result of model fitting in the multivariate meta-regression analysis was the difference in the risk of new vertebral fracture among the study populations with different prevalence of vertebral fracture.

The present results of the subgroup analysis showed a significant interaction between the proportion of subjects with prevalent vertebral fracture and the percent change in lumbar spine BMD from baseline at 3 years, and the regression coefficient was lower in the subgroup of higher tertile. It indicates that the correlation between the change in BMD and the incidence of new vertebral fracture is different between the study populations with high and low prevalence of vertebral fracture; the higher prevalence of vertebral fracture the study group has, the greater effect of increase in lumbar spine BMD on the prevention of new vertebral fracture would be observed. The degree of prevalence of vertebral fracture in the population should be considered when the association between

change in lumbar spine BMD and incidence of vertebral fracture is examined.

4. Overall Discussion

Considering the results of the univariate meta-regression analysis in Research 1, we suggest that the ethnic/regional difference should be considered as one of the important factors that influence the incidence of new vertebral fracture, a primary endpoint of osteoporosis study, when the Caucasian reference range is used in clinical trials. We expect that accumulation of epidemiology studies in each region would contribute to the establishment of a reference range in each region. And, it would be helpful to decrease the variability of incidence of fractures causing by ethnic difference.

The present study showed that prevalent fracture was the most important factor influencing the incidence of new fracture and that the prediction of fracture risk by change in lumbar spine BMD was improved by the adjustment with the proportion of subjects with prevalent vertebral fracture in the study population. It indicated that it would be possible to evaluate drug efficacy by fracture risk estimation model based on the change in BMD adjusted by other factors such as prevalent fracture in future osteoporosis drug development. We expect continuous discussions on the development of better fracture risk estimation model to decrease the burden of osteoporosis drug development.

The present study has several limitations. First, the analyses in the present study were not performed at the patient level but instead used summary data; therefore, accurate

assessment may be lacking as the nature of meta-analysis. However, the direction of the coefficients for each factor in both the univariate and multivariate meta-regression analysis were consistent with the previous epidemiological report. Second, this study used data from RCTs. The characteristics of subjects who participate in clinical trials of new therapy may have affected the generalizability of this study result. Third, several recent clinical trials for osteoporosis drugs such as Romosozumab and Odanacatib, which showed drastic increase in BMD, were not included in this study because of the short study period and/or undisclosed study results. Further investigations including the studies which have big change in BMD in a short study period are expected. Fourth, the present study could not directly compare the ethnic difference based on the data of proportion of Asian, Black, and Hispanic populations. Further investigation is needed to reveal the effect of each ethnic population on the incidence of new vertebral fracture.

5. Conclusion

The result of Research 1 showed that prevalent vertebral fracture was the most important factor to predict subsequent vertebral fracture. In Research 2, the change in lumbar spine BMD, not BMD T-score at one time point, was shown to have a significant correlation with the incidence of vertebral fracture. The prediction of fracture risk by change in lumbar spine BMD was improved by the adjustment with the proportion of subjects with prevalent vertebral fracture in the study population. The degree of

prevalence of vertebral fracture in the population should be considered when the association between change in lumbar spine BMD and incidence of vertebral fracture is examined. We also suggest that it would be possible to evaluate drug efficacy by fracture risk estimation model based on the change in BMD adjusted by other factors such as prevalent fracture in future osteoporosis drug development.

References

1. Burge R, Dawson-Hughes B, Solomon DH, Wong JB, King A, Tosteson A. Incidence and economic burden of osteoporosis-related fractures in the United States, 2005-2025. J Bone Miner Res. 2007;22(3):465-75.
2. Akesson K, Woolf AD. Bone: Risk of death persists for years after hip fracture. Nat Rev Rheumatol. 2010;6(10):557-8.
3. Kanis JA, Johnell O, De Laet C, Johansson H, Oden A, Delmas P, et al. A meta-analysis of previous fracture and subsequent fracture risk. Bone. 2004;35(2):375-82.
4. Schousboe JT, Fink HA, Taylor BC, Stone KL, Hillier TA, Nevitt MC, et al. Association between self-reported prior wrist fractures and risk of subsequent hip and radiographic vertebral fractures in older women: a prospective study. J Bone Miner Res. 2005;20(1):100-6.
5. FDA. Guidelines for preclinical and clinical evaluation of agents used in the prevention or treatment of postmenopausal osteoporosis. Division of Metabolism and Endocrine Drug Products, Rockville, MD. 1994.
6. Committee for Medicinal Products for Human Use.. Guideline on the evaluation of medicinal products in the treatment of primary osteoporosis. London, Doc. Ref: CPMP/EWP/552/95 Rev. 2; 2006.
7. Madrasi K, Li F, Kim MJ, Samant S, Voss S, Kehoe T, et al. Regulatory Perspectives in Pharmacometric Models of Osteoporosis. J Clin Pharmacol. 2018;58(5):572-85.
8. Johnell O, Kanis JA. An estimate of the worldwide prevalence and disability associated with osteoporotic fractures. Osteoporos Int. 2006;17(12):1726-33.
9. Bow CH, Cheung E, Cheung CL, Xiao SM, Loong C, Soong C, et al. Ethnic difference of clinical vertebral fracture risk. Osteoporos Int. 2012;23(3):879-85.
10. Cauley JA, Palermo L, Vogt M, Ensrud KE, Ewing S, Hochberg M, et al. Prevalent vertebral fractures in black women and white women. J Bone Miner Res. 2008;23(9):1458-67.
11. Kanis JA on behalf of WHO Scientific Group. Assessment of osteoporosis at the primary health care level. Technical Report. World Health Organization Collaborating Centre for Metabolic Bone Diseases. UK: University of Sheffield;

2007.
12. Schousboe JT, Shepherd JA, Bilezikian JP, Baim S. Executive summary of the 2013 International Society for Clinical Densitometry Position Development Conference on bone densitometry. J Clin Densitom. 2013;16(4):455-66.
13. Kanis JA, Gluer CC. An update on the diagnosis and assessment of osteoporosis with densitometry. Committee of Scientific Advisors, International Osteoporosis Foundation. Osteoporos Int. 2000;11(3):192-202.
14. Soen S, Fukunaga M, Sugimoto T, Sone T, Fujiwara S, Endo N, et al. Diagnostic criteria for primary osteoporosis: year 2012 revision. J Bone Miner Metab. 2013;31(3):247-57.
15. Cummings SR, Black DM, Thompson DE, Applegate WB, Barrett-Connor E, Musliner TA, et al. Effect of alendronate on risk of fracture in women with low bone density but without vertebral fractures: results from the Fracture Intervention Trial. Jama. 1998;280(24):2077-82.
16. Cosman F, Crittenden DB, Ferrari S, Lewiecki EM, Jaller-Raad J, Zerbini C, et al. Romosozumab FRAME Study: A Post Hoc Analysis of the Role of Regional Background Fracture Risk on Nonvertebral Fracture Outcome. J Bone Miner Res. 2018.
17. Moher D, Shamseer L, Clarke M, Ghersi D, Liberati A, Petticrew M, et al. Preferred reporting items for systematic review and meta-analysis protocols (PRISMA-P) 2015 statement. Systematic Reviews. 2015;4(1):1.
18. Jadad AR, Moore RA, Carroll D, Jenkinson C, Reynolds DJ, Gavaghan DJ, et al. Assessing the quality of reports of randomized clinical trials: is blinding necessary? Control Clin Trials. 1996;17(1):1-12.
19. Team RC. R: A language and enviroment for statistical computing. Vienna, Austria: R foundation for statistical computing; 2017.
20. Harris ST, Watts NB, Jackson RD, Genant HK, Wasnich RD, Ross P, et al. Four-year study of intermittent cyclic etidronate treatment of postmenopausal osteoporosis: three years of blinded therapy followed by one year of open therapy. Am J Med. 1993;95(6):557-67.
21. Liberman UA, Weiss SR, Broll J, Minne HW, Quan H, Bell NH, et al. Effect of oral alendronate on bone mineral density and the incidence of fractures in

postmenopausal osteoporosis. The Alendronate Phase III Osteoporosis Treatment Study Group. N Engl J Med. 1995;333(22):1437-43.

22. Black DM, Cummings SR, Karpf DB, Cauley JA, Thompson DE, Nevitt MC, et al. Randomised trial of effect of alendronate on risk of fracture in women with existing vertebral fractures. Fracture Intervention Trial Research Group. Lancet. 1996;348(9041):1535-41.
23. Harris ST, Watts NB, Genant HK, McKeever CD, Hangartner T, Keller M, et al. Effects of risedronate treatment on vertebral and nonvertebral fractures in women with postmenopausal osteoporosis: a randomized controlled trial. Vertebral Efficacy With Risedronate Therapy (VERT) Study Group. Jama. 1999;282(14):1344-52.
24. Ettinger B, Black DM, Mitlak BH, Knickerbocker RK, Nickelsen T, Genant HK, et al. Reduction of vertebral fracture risk in postmenopausal women with osteoporosis treated with raloxifene: results from a 3-year randomized clinical trial. Multiple Outcomes of Raloxifene Evaluation (MORE) Investigators. Jama. 1999;282(7):637-45.
25. Reginster J, Minne HW, Sorensen OH, Hooper M, Roux C, Brandi ML, et al. Randomized trial of the effects of risedronate on vertebral fractures in women with established postmenopausal osteoporosis. Vertebral Efficacy with Risedronate Therapy (VERT) Study Group. Osteoporos Int. 2000;11(1):83-91.
26. Chesnut CH, 3rd, Silverman S, Andriano K, Genant H, Gimona A, Harris S, et al. A randomized trial of nasal spray salmon calcitonin in postmenopausal women with established osteoporosis: the prevent recurrence of osteoporotic fractures study. PROOF Study Group. Am J Med. 2000;109(4):267-76.
27. Reginster JY, Christiansen C, Roux C, Fechtenbaum J, Rouillon A, Tou KP. Intermittent cyclic tiludronate in the treatment of osteoporosis. Osteoporos Int. 2001;12(3):169-77.
28. Alexandersen P, Toussaint A, Christiansen C, Devogelaer JP, Roux C, Fechtenbaum J, et al. Ipriflavone in the treatment of postmenopausal osteoporosis: a randomized controlled trial. Jama. 2001;285(11):1482-8.
29. Chesnut CH, 3rd, Skag A, Christiansen C, Recker R, Stakkestad JA, Hoiseth A, et al. Effects of oral ibandronate administered daily or intermittently on fracture risk in postmenopausal osteoporosis. J Bone Miner Res. 2004;19(8):1241-9.

30. Recker R, Stakkestad JA, Chesnut CH, 3rd, Christiansen C, Skag A, Hoiseth A, et al. Insufficiently dosed intravenous ibandronate injections are associated with suboptimal antifracture efficacy in postmenopausal osteoporosis. Bone. 2004;34(5):890-9.
31. Meunier PJ, Roux C, Seeman E, Ortolani S, Badurski JE, Spector TD, et al. The effects of strontium ranelate on the risk of vertebral fracture in women with postmenopausal osteoporosis. N Engl J Med. 2004;350(5):459-68.
32. Reginster JY, Seeman E, De Vernejoul MC, Adami S, Compston J, Phenekos C, et al. Strontium ranelate reduces the risk of nonvertebral fractures in postmenopausal women with osteoporosis: Treatment of Peripheral Osteoporosis (TROPOS) study. J Clin Endocrinol Metab. 2005;90(5):2816-22.
33. Black DM, Delmas PD, Eastell R, Reid IR, Boonen S, Cauley JA, et al. Once-yearly zoledronic acid for treatment of postmenopausal osteoporosis. N Engl J Med. 2007;356(18):1809-22.
34. Silverman SL, Christiansen C, Genant HK, Vukicevic S, Zanchetta JR, de Villiers TJ, et al. Efficacy of bazedoxifene in reducing new vertebral fracture risk in postmenopausal women with osteoporosis: results from a 3-year, randomized, placebo-, and active-controlled clinical trial. J Bone Miner Res. 2008;23(12):1923-34.
35. Cummings SR, Ettinger B, Delmas PD, Kenemans P, Stathopoulos V, Verweij P, et al. The effects of tibolone in older postmenopausal women. N Engl J Med. 2008;359(7):697-708.
36. Cummings SR, San Martin J, McClung MR, Siris ES, Eastell R, Reid IR, et al. Denosumab for prevention of fractures in postmenopausal women with osteoporosis. N Engl J Med. 2009;361(8):756-65.
37. Cummings SR, Ensrud K, Delmas PD, LaCroix AZ, Vukicevic S, Reid DM, et al. Lasofoxifene in postmenopausal women with osteoporosis. N Engl J Med. 2010;362(8):686-96.
38. Cummings SR, McClung M, Reginster JY, Cox D, Mitlak B, Stock J, et al. Arzoxifene for prevention of fractures and invasive breast cancer in postmenopausal women. J Bone Miner Res. 2011;26(2):397-404.
39. Henriksen K, Byrjalsen I, Andersen JR, Bihlet AR, Russo LA, Alexandersen P, et al.

A randomized, double-blind, multicenter, placebo-controlled study to evaluate the efficacy and safety of oral salmon calcitonin in the treatment of osteoporosis in postmenopausal women taking calcium and vitamin D. Bone. 2016;91:122-9.

40. Bruyere O, Roux C, Detilleux J, Slosman DO, Spector TD, Fardellone P, et al. Relationship between bone mineral density changes and fracture risk reduction in patients treated with strontium ranelate. J Clin Endocrinol Metab. 2007;92(8):3076-81.
41. Blake GM, Fogelman I. Long-term effect of strontium ranelate treatment on BMD. J Bone Miner Res. 2005;20(11):1901-4.
42. Nielsen SP, Slosman D, Sorensen OH, Basse-Cathalinat B, De Cassin P, Roux CR, et al. Influence of strontium on bone mineral density and bone mineral content measurements by dual X-ray absorptiometry. J Clin Densitom. 1999;2(4):371-9.
43. Hochberg MC, Ross PD, Black D, Cummings SR, Genant HK, Nevitt MC, et al. Larger increases in bone mineral density during alendronate therapy are associated with a lower risk of new vertebral fractures in women with postmenopausal osteoporosis. Fracture Intervention Trial Research Group. Arthritis Rheum. 1999;42(6):1246-54.
44. Wasnich RD, Miller PD. Antifracture efficacy of antiresorptive agents are related to changes in bone density. J Clin Endocrinol Metab. 2000;85(1):231-6.
45. Kobayashi T, Kaneko M, Narukawa M. Investigation of the Influencing Factors on the Incidence of Vertebral Fracture:: A Meta-analysis Based on Placebo Data of Randomized Clinical Trials. Ther Innov Regul Sci. 2019:2168479019833657.
46. Cooper C, O'Neill T, Silman A. The epidemiology of vertebral fractures. European Vertebral Osteoporosis Study Group. Bone. 1993;14 Suppl 1:S89-97.
47. Pouilles JM, Tremollieres F, Louvet JP, Fournie B, Morlock G, Ribot C. Sensitivity of dual-photon absorptiometry in spinal osteoporosis. Calcif Tissue Int. 1988;43(6):329-34.
48. ISCD. 2015 ISCD Official Positions – Adult. 2015 https://www.iscd.org/official-positions/2015-iscd-official-positions-adult/ Accessed 17 Oct 2019.

Acknowledgement

I appreciate to the advice and suggestion to my research by Professor Mamoru Narukawa and associate professor Masayuki Kaneko and kind supports by members in our laboratory.

I would like to thank Professor Yukio Suzuki, Professor Nobuhiko Okada, and Professor Yuji Yoshiyama for their reviews and helpful discussion on this study.