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Vitamin B6 as a novel risk biomarker of fractured ankles

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Abstract

Ankle fractures are the most common intra-articular fractures. Osteoporosis is a common and frequent disease among the elderly with a poor prognosis and high risk of fractured ankles. However, the relationship between vitamin B6 and the incidence of fractured ankles in patients with osteoporosis is unclear.

A total of 101 patients with osteoporosis were recruited. Clinical and followed-up information was recorded. And the vitamin B6, albumin, globulin, and hemoglobin in the blood were tested. Pearson's chi-squared and spearman test were performed to analyze the correlation between fractured ankles and relative parameters. Univariate and multivariate logistic regression, receiver operating characteristic curve analysis, univariate and multivariate Cox proportional hazards regression analysis, and Kaplan–Meier method were also performed.

There exist strong relation between the expression level of vitamin B6 and fractured ankle ($P < .001$). The expression of vitamin B6 [Odd ratio (OR) = 12.071, 95% confidence interval (CI): 4.69–31.143, $P < .001$] has a clear correlation with whether the patients have fractured ankles via the univariate logistic regression analysis. In terms of multivariate logistic regression level, vitamin B6 (OR = 15.384, 95% CI:5.195–45.556, $P < .001$) was significantly associated with fractured ankle. In addition, expression level of vitamin B6 [hazard ratio (HR) = 11.684, 95% CI: 6.419–21.267, $P < .001$] was significantly associated with Maintenance time from recovery to recurrence (MRTT) of patients with osteoporosis.

Enhanced vitamin B6 is significantly correlated with the poor prognosis of patients with osteoporosis and the increasing incidence of fractured ankles.

Abbreviations: AUC = receiver operating characteristic, CI = confidence interval, ER = estrogen receptor, ER-β = estrogen receptor β, ER-α= estrogen receptor α, HR = hazard ratio, HRT = hormone replacement therapy, IGF-1 = insulin-like growth factor-1, MTRR = Maintenance time from recovery to recurrence, NYHA = New York Heart Association, OR = Odd ratio, PLP = Pyridoxal 5-phosphate, ROC = receiver operating characteristic.

Keywords: estrogen, fractured ankles, osteoporosis, prognosis, vitamin B6

1. Introduction

Osteoporosis is a common and frequent disease among the elderly. As the disease progresses, bone cell-mediated bone remodeling would be emerging, with reducing bone mass, damaging bone microstructure, and leading to osteoporosis.[1] With the aggravation of the current aging society, the incidence of osteoporosis in the elderly is also increasing year by year.[2] Due to the deterioration of physical movement of the elderly, walking is unstable, and patients are prone to falls or collisions, and often accompanied by fractured ankle. In recent years, despite advances in surgical techniques and molecular targeted therapy and immunotherapy, the prognosis of osteoporosis is still unsatisfactory.[3,4]

Vitamin B6 [also known as Pyridoxal 5-phosphate (PLP)] is an essential cofactor involved in the metabolism of homocysteine and the synthesis of catecholamine and other neurotransmitters.[5] PLP is an important nutrient essential for maintaining normal physiological metabolic activities of the body.[6] Vitamin B6 is involved in the synthesis and metabolism of amino acids, glycogen, neurotransmitters, sphingolipids, heme, and nucleic acids to promote the normal function of the nervous and skeletal muscle systems.[7] Studies have pointed out that the body nutrients, amino acid metabolism and vitamin B6 are inseparable.[8] During disease process of osteoporosis, osteoporosis tissues need to constantly absorb glucose, Vitamins, amino acid and other nutrient from the body.[9] In a cross-sectional study by

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The datasets generated during and/or analyzed during the present study are available from the corresponding author on reasonable request.

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Wang et al, the association between serum vitamin B6 concentration and risk of osteoporosis in the middle-aged and older people (n = 1829) in China was investigated. Wang et al indicates that a relatively low serum vitamins B6 concentration is a risk factor for osteoporosis in postmenopausal women.[10] Meyer et al investigated that combined high intake of vitamins B6 and B12 was associated with an increased risk of hip fracture.[11] However, the relationship between vitamin B6 and the incidence of fractured ankles in patients with osteoporosis is unclear.

Therefore, this study hypothesized that during the occurrence and development of osteoporosis, the increased vitamin B6 would increase the risk of fractured ankles. Based on the above hypothesis, 101 patients with osteoporosis were recruited, and correlation analysis, and receiver operating characteristic curve analysis were used to explore the relationship and risk value of the expression of vitamin B6 for the prognosis of osteoporosis and the occurrence of fractured ankles. The results might reveal vitamin B6 as a new target for the osteoporosis, and provide new ideas for the molecular mechanism of osteoporosis occurrence and development.

2. Methods

2.1. Patients and ethics

A total of 101 patients diagnosed as osteoporosis with (or without fractured ankles) were recruited at the Second Affiliated Hospital of Luohe Medical College, from March 2015 to June 2020.

Interpretation of the sample size of 101 people:

$$N = Z_{\alpha}[2P_{mean}(1-P_{mean})]^{0.5} + Z_{\beta}[P_1(1-P_1)+P_2(1-P_2)]^{0.5^2}/(P_1-P_2)^2$$

N represents the sample size of one group, P_1 represents the incidence of ankle fracture in the experimental group and P_2 represents the incidence of ankle fracture in the control group, $P_{mean} = (P_1 + P_2)/2$, Z_{α}= The standard normal difference at the α level; Z_{β}= The standard normal difference at the beta level. According to literature studies, the incidence of foot and ankle fractures is about 10%. α=0.01, β=0.05. This study is a bilateral test, and P_1 is set as 0.4 and P_2 is set as 0.25.

In addition, considering the loss of follow-up rate of 10% and the number of patients in the bidders and partners, 101 subjects are planned to be included in this study.

Inclusion criteria: 18 < age < 80 years old; diagnosed as osteoporosis; normal cardiopulmonary function; normal clotting function.

Exclusion criteria: age≤18, or ≥80; Patients and their families did not agree to participate in the trial; patients who had taken vitamin B6 supplements; Heart failure, New York Heart Association (NYHA) standards III or grade; Patients with hepatitis B, hepatitis C, syphilis, Acquired Immune Deficiency Syndrome and other infectious diseases; patients with unstable vital signs.

This study was approved by the Ethics Committee of the Second Affiliated Hospital of Luohe Medical College. Written informed consent was obtained from all patients.

2.2. Parameters in the research

Based clinical information of patients were classed according to sex (Male/Female), age (≤65/>65), albumin (Low/High), globulin (Low/High), fractured ankle (No/Yes), and hemoglobin (Low/High). And the patients were followed up five years, and the MTRR was recorded. MTRR represents the maintenance time from March 2015 recovery to June 2020 relapse. The albumin <50 g/L was defined as low, and albumin >50 g/L was defined as high. The globulin <25 g/L was defined as low, and a globulin >25 g/L was defined as high. Yes of fractured ankle: X-ray showed a marked fracture line at the ankle; No of fractured ankle: X-ray didn't show a marked fracture line at the ankle. A hemoglobin <135 g/L was defined as low, and a hemoglobin >135 g/L was defined as high.

2.3. The detection of relative blood parameters

Venous blood samples were immediately sent for examination and then tested by tyrosine decarboxylase for vitamin B6 level. The level of albumin and globulin in the blood was tested according to the instructions for albumin and globulin test kit (Beijing Lidan Biochemical Technology Co., Ltd, Beijing, China). The hemoglobin level was detected by routine blood test.

2.4. Statistical methods

The data was expressed as percentage of total. By using the Pearson's chi-squared test, associations between the clinical parameters and vitamin B6 were analyzed. The Spearman–rho test was executed to compare clinical data and vitamin B6 for the correlation analysis. Univariate and multivariate logistic regression analysis was used to calculate the OR of each variable for fractured ankle. A receiver operating characteristic (ROC) curve analysis was performed to determine the ability of the clinical parameters to predict the fractured ankle. By using univariate and multivariate Cox proportional hazards regression analysis, potential prognostic factors for MTRR were explored. The further illustrating for MTRR was made by the Kaplan–Meier method.

All statistical analyses were conducted using SPSS software, version 21.0 (IBM Corp., Armonk, NY, USA). A P-value < .05 was considered statistically significant.

3. Results

3.1. Strong associations between fractured ankle and vitamin B6 based on χ^2 test

Table 1 summarized the associations between vitamin B6 and the related clinical factors according to the pearson's chi-squared test. Among the individuals, fractured ankle ($P < .001$) and hemoglobin ($P = .041$) were markedly related to the vitamin B6. However, no significant associations were found between sex ($P = .687$), age ($P = .326$), albumin ($P = .820$), globulin ($P = .431$) and vitamin B6. (Table 1)

3.2. Further associations between fractured ankle and vitamin B6 by Spearman's correlation test

To confirm whether the potentially characteristics about fractured ankle and hemoglobin factors played an important role on vitamin B6, a further correlation analysis was performed. Spearman's correlation coefficient displayed that vitamin B6 were significantly correlated with the fractured ankle ($\rho = 0.548$, $P < .001$), and hemoglobin ($\rho = 0.203$, $P = .042$). However, there

Table 1

Relevant characteristics of patients with Vitamin B6.

Parameters			Vitamin B6		
			Low (%)	High (%)	*P*
Sex	Male	61	28 (27.7%)	33 (32.7%)	.687
	Female	40	20 (19.8%)	20 (19.8%)	
Age	≤65	58	30 (29.7%)	28 (27.7%)	.326
	>65	43	18 (17.8%)	25 (24.8%)	
Albumin	Low	43	21 (20.8%)	22 (21.8%)	.820
	High	58	27 (26.7%)	31 (30.7%)	
Globulin	Low	65	29 (28.7%)	36 (35.6%)	.431
	High	36	19 (18.8%)	17 (16.8%)	
Fractured ankles*	No	53	39 (38.6%)	14 (13.9%)	<.001*
	Yes	48	9 (8.9%)	39 (38.6%)	
Hemoglobin*	Low	34	21 (20.8%)	13 (12.9%)	.041*
	High	67	27 (26.7%)	40 (39.6%)	

Pearson's chi-squared test was used.
* $P<.05$.

was no significant correlation between sex (ρ=-0.040, $P=.690$), age (ρ=0.098, $P=.331$), albumin (ρ=0.023, $P=.822$), globulin (ρ=-0.078, $P=.436$) and vitamin B6. (Table 2)

3.3. Univariate logistic regression analysis of odds ratios between fractured ankle and correlative factors

In addition, our study used binary logistic regression (including univariate logistic regression and multivariate logistic regression) to determine the association between correlative parameters and fractured ankle, odds ratios (ORs) and 95% confidence intervals (95% CIs), in order to further determine the risk factors and risk groups of fractured ankles. Table 3 describes the ORs and 95% CI of the study subjects at the univariate level using univariate logistic regression and concludes that the expression of vitamin B6 (OR= 12.071, 95% CI: 4.69–31.143, $P<.001$) and hemoglobin (OR= 3.145, 95% CI: 1.302–7.596, $P=.011$) have a clear correlation with whether the patients have fractured ankles. (Table 3)

3.4. Multivariate logistic regression analysis for correlative factors and fractured ankle

Table 4 applied multivariate logistic regression to describe the OR and 95% CI of the study subjects at the multivariate level. In terms of multivariate logistic regression level, vitamin B6 (OR= 15.384, 95% CI:5.195–45.556, $P<.001$) was significantly associated with fractured ankle, whereas sex (OR=2.116, 95% CI:0.724-6.186, $P=.0.171$), age (OR=0.423, 95% CI:0.147–1.219, $P=.111$), albumin (OR=2.001, 95% CI:0.645–6.208, $P=.230$), globulin (OR=0.666, 95% CI:0.218–2.032, $P=.0.475$) and hemoglobin (OR=2.244, 95% CI: 0.771–6.538, $P=.138$) showed no significant associations with fractures of ankle. (Table 4)

3.5. Vitamin B6 could sensitively and specifically predict fractured ankle through the ROC curve

Via the ROC curve analysis, the sex, age, albumin, and globulin couldn't predict fractured ankle sensitively and specifically ($P>.005$, Fig. 1A-D). The vitamin B6 was most associated with fractured ankle (area under the curve (AUC)=0.774; $P<.001$) (Fig. 1E). The hemoglobin was most closely related to a greater risk of fractured ankle (AUC=0.622; $P=.034$) (Fig. 1F).

3.6. Univariate cox regression for the proportional hazard analysis of vitamin B6

Table 5 presented the univariate HR and 95% CI for patients who underwent osteoporosis. For overall survival, subjects who

Table 2

The relationship between characteristics of patients and Vitamin B6.

Characteristics	Vitamin B6	
	ρ	*P*
Sex	−0.040	.690
Age	0.098	.331
Albumin	0.023	.822
Globulin	−0.078	.436
Fractured ankles*	0.548	<.001*
Hemoglobin*	0.203	.042*

Spearman correlation test was used.
* $P<.05$.

Table 3

Correlative parameters' effect on fractured ankles based on univariate logistic proportional regression analysis.

Parameters		Fractured ankles		
		OR	95% CI	*P*
Sex	Male	1		.224
	Female	1.645	0.737–3.674	
Age	≤65	1		.563
	>65	0.792	0.359–1.748	
Albumin	Low	1		.168
	High	1.756	0.789–3.908	
Globulin	Low	1		.645
	High	0.825	0.364–1.869	
Vitamin B6*	Low	1		<.001*
	High	12.071	4.69–31.143	
Hemoglobin*	Low	1		.011*
	High	3.145	1.302–7.596	

95% CI=95% confidence interval, OR=odds ratio.
* $P<.05$.

Table 4

The characteristics and their effect on fractured ankles based on multivariate Logistic proportional regression analysis.

	Fractured ankles		
Characteristics	**OR**	**95%CI**	***P***
Sex	2.116	0.724–6.186	.171
Age	0.423	0.147–1.219	.111
Albumin	2.001	0.645–6.208	.230
Globulin	0.666	0.218–2.032	.475
Vitamin B6*	15.384	5.195–45.556	<.001*
Hemoglobin	2.244	0.771–6.538	.138

95% CI = 95% confidence interval, OR = odds ratio.
* $P < .05$.

had high vitamin B6 had obviously lower MTRR than subjects who had low vitamin B6 level, and the HR is 8.799 (95% CI, 5.146–15.044, $P < .001$). However, sex (HR = 0.806, 95% CI: 0.521–1.248, $P = .334$), age (HR = 1.230, 95% CI: 0.804–1.880, $P = .340$), albumin (HR = 0.788, 95% CI: 0.501–1.237, $P = .300$), globulin (HR = 1.247, 95% CI: 0.794–1.959, $P = .337$) and hemoglobin (HR = 1.034, 95% CI: 0.655–1.632, $P = .885$) had no disadvantageous for MTRR significantly. (Table 5)

3.7. Analysis of MTRR based on multivariate cox regression for the proportional hazards of related characteristics

In order to effectively control the influence of confounding factors, all factors were incorporated into the multivariate cox regression model simultaneously. Table 6 showed the result of multivariate cox proportional regression analysis, vitamin B6 gene level (HR = 11.684, 95% CI: 6.419–21.267, $P < .001$) was significantly associated with MTRR. However, sex (HR = 0.754, 95% CI: 0.469–1.211, $P = .243$), age (HR = 1.020, 95% CI: 0.633–1.644, $P = .934$), albumin (HR = 0.680, 95% CI: 0.422–1.096, $P = .113$), globulin (HR = 1.639, 95% CI: 0.999–2.692, $P = .051$) and hemoglobin (HR = 0.738, 95% CI: 0.488–1.216, $P = .233$) have no significant correlation with MTRR. (Table 6)

Based on the further Kaplan–Meier method, sex, age, albumin, globulin, and hemoglobin were not related with the overall survival of patients with osteoporosis. However, the higher vitamin B6 expression was, the shorter MTRR was. (Fig. 2)

4. Discussion

Based on the Pearson χ^2 and Spearman correlation tests, there exist strong relation between the expression level of vitamin B6 and fractured ankle. The expression of vitamin B6 has a clear correlation with fractured ankles via the univariate and multivariate logistic regression analysis. In addition, expression level of vitamin B6 was significantly associated with MTRR of patients with osteoporosis. Therefore, in this study, we discarded the old indicators and explored the relationship between vitamin B6, a novel indicator, and ankle fractures in patients with osteoporosis.

Vitamin B6, also known as pyridoxine, includes pyridoxol, pyridoxal and pyridoxine. It exists in the form of phosphate in the body.[12] Vitamin B6 is a component of some coenzymes in human body, which is involved in a variety of metabolic reactions, especially the metabolism of amino acids.[13] Vitamin B6 plays an important role in protein metabolism.[14] Therefore, when the expression level of vitamin B6 gene increases, it promotes the production of vitamin B6, enhances the metabolism of protein, amino acid and glucose, and thus enhances the supply of nutrients in the body, which could provide energy for the bone remodeling of osteoporosis.[15] Furthermore, the rapid bone remodeling with an abundant nutrient supply further damages and erodes bone tissue, reduces bone mass, leads to brittle bone, and thus increases the clinical risk of fractured ankles.[16] Dai et al indicates that there was a significant inverse relationship between dietary vitamins B6 intake and hip fracture risk among women.[17] Our study suggested that enhanced vitamin B6 is significantly correlated with the poor prognosis of patients with osteoporosis and the increasing incidence of fractured ankles, and our conclusion is similar to Dai et al. However, a larger sample size was needed to verify our conclusion.

In addition, studies have shown that vitamin B6 is also involved in estrogen metabolism, reducing estrogen activity and possibly reducing estrogen levels in the body.[18] Estrogen is one of the main hormones regulating bone metabolism.[19] The deficiency of estrogen will increase bone cell-mediated bone remodeling, reduce bone mass, damage bone microstructure and lead to osteoporosis.[20] Estrogen is an important endocrine hormone in the body, which plays an important role in growth and development by binding to the corresponding estrogen receptor.[21] In the process of bone growth and development, estrogen regulates bone formation in two main ways: indirectly promoting bone growth by stimulating growth hormone/insulin-like growth factor-1 (IGF-1) axis; Binding to bone surface estrogen receptor (ER) directly affects bone metabolism, which is an important way to regulate bone metabolism.[22] The current clinical use of hormone replacement therapy (HRT) in postmenopausal women with osteoporosis is likely to be related to the above approach. Some studies have suggested that the occurrence of osteoporosis is related to the estrogen receptor in bone tissue.[23] Estrogen receptor is a glycoprotein that can specifically bind with estrogen, located in the cytoplasm and nucleus.[24] When estrogen binds with estrogen, it activates related pathways to regulate biological activity. There are two subtypes of estrogen receptor: estrogen receptor α (ER-α) and estrogen receptor β (ER-β), which are mostly distributed in cancellous bone and rarely distributed in the bone cortex.[25] Therefore, the effect of estrogen level decline on bone trabecula is greater than that on bone cortex. The transcription efficiency of ER-α during human development is significantly higher than that of ER-β, so the effect of estrogen on bone through ER-α may be greater than that of ER-β on bone.[26] Therefore, in the presence of increased vitamin B6, increased vitamin B6 might lead to decreased estrogen levels, which in turn increases the incidence of osteoporosis and the risk of ankle fractures. In a longitudinal follow-up study of the Framingham Osteoporosis Study,[10] bone loss in older adults was inversely associated with serum Vitamin B6 concentrations, with 22% of participants diagnosed with a serum Vitamin B6 deficiency. The Becker muscular dystrophy of the femoral neck in the patients with serum Vitamin B6 deficiency was lower than that in the patients with normal serum Vitamin B6. Therefore, Vitamin B6 deficiency may be a risk factor for bone loss.

The Rotterdam study also showed that fracture risk decreased with increased vitamin B6 intake, which was not associated with bone mineral density.[10] In the Framingham Osteoporosis Study, low plasma vitamin B6 concentrations were associated with a

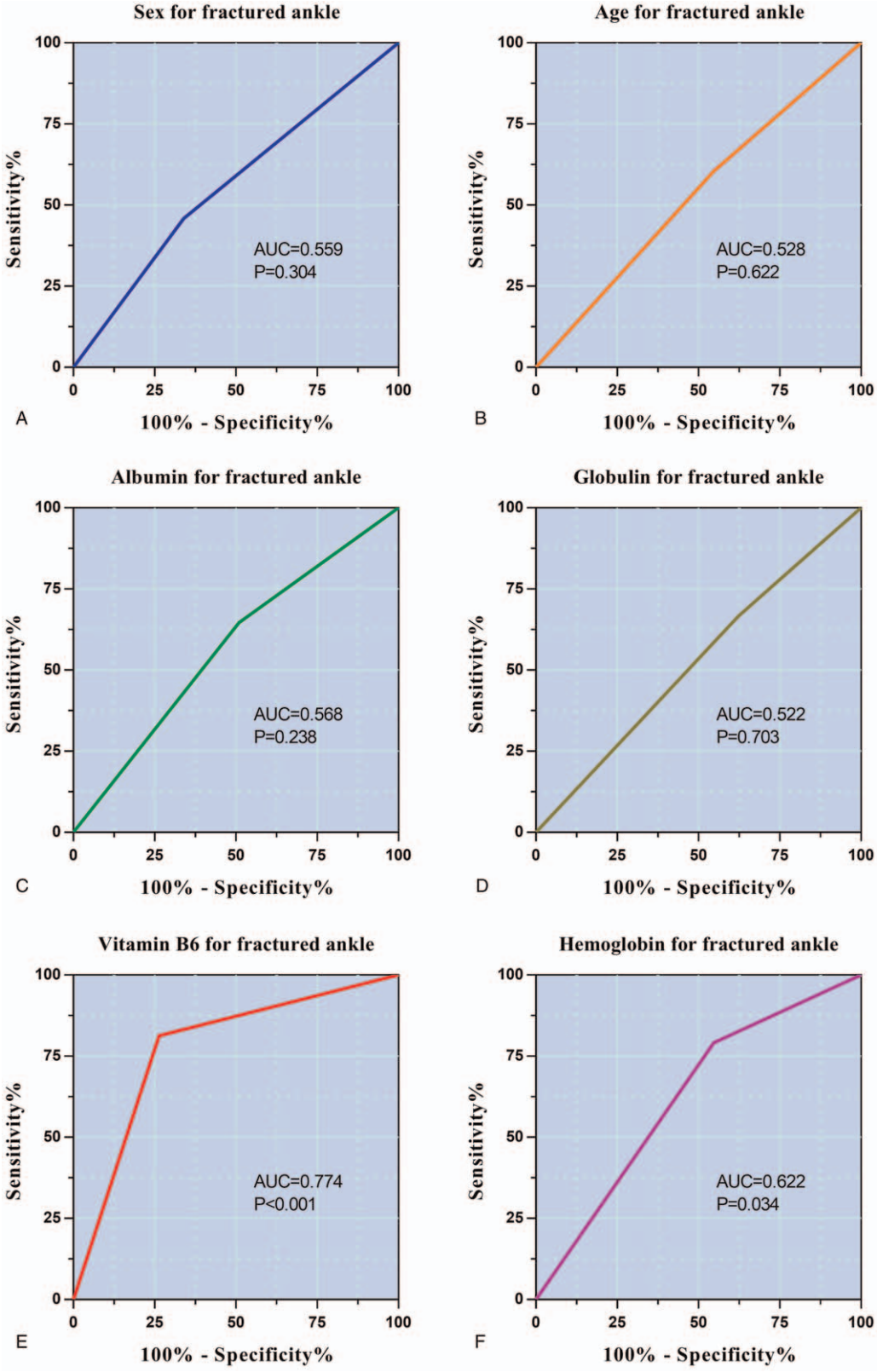

Figure 1. ROC curve of the predictive value between clinical parameters and fractured ankles. (A) sex, (B) age, (C) albumin, (D) globulin, (E) vitamin B6, (F) hemoglobin.

Table 5

Characteristics and their effect on MTRR based on univariate Cox proportional regression analysis.

Characteristics			MTRR		
			HR	95% CI	P
	Male	61	1		.334
	Female	40	0.806	0.521–1.248	
Age	≤65	58	1		.340
	>65	43	1.230	0.804–1.880	
Albumin	Low	43	1		.300
	High	58	0.788	0.501–1.237	
Globulin	Low	65	1		.337
	High	36	1.247	0.794–1.959	
Vitamin B6*	Low	53	1		<.001*
	High	48	8.799	5.146–15.044	
Hemoglobin	Low	34	1		.885
	High	67	1.034	0.655–1.632	

95% CI = 95% confidence interval, HR = hazard ratio, MTRR = Maintenance time from recovery to recurrence.
* $P < .05$.

higher risk of fractures in older adults. In addition, another study confirmed that in women, dietary pyridoxine intake and risk of hip fracture was significantly negative related bone mineral density and bone are the key factors that determine bone strength, and the collagen/linked to maintain bone plays an important role in vitamin B6 is necessary for collagen cross-linking of lysine oxidase, vitamin B6 deficiency can reduce the concentration of cross-linking intermediates, damages the formation of collagen cross-links in bones, resulting in poor bone quality. This may be one reason for the association between high fracture risk and low vitamin B6 intake or low circulating vitamin B6 concentration.

The possible mechanism by which excess vitamin B6 exposure increases the risk of falls through neurological symptoms may particularly increase the risk of fractures in women with lower body mass index, who are more prone to hip fractures when they fall.[11] This research demonstrated our results.

The role of B vitamin supplements in fracture prevention and bone health makes sense. Previous studies have shown that hyperhomocysteine may interfere with collagen cross-linking. Collagen cross-linking plays an important role in determining the stability and strength of the collagen network, and if collagen is lacking, the bone matrix may be damaged, leading to an increased risk of fracture. In addition, homocysteine may promote oxidative damage, leading to increased fractures. For example, cytotoxic reactive oxygen species (including superoxide anion and hydroxyl radicals) are produced when homocysteine is oxidized to produce homocysteine and homocysteine thionolide, which in turn trigger lipid peroxidation.[27]

Table 6

Characteristics and their effect on MTRR based on multivariate Cox regression analysis.

Characteristics	MTRR		
	HR	95% CI	P
Sex	0.754	0.469–1.211	.243
Age	1.020	0.633–1.644	.934
Albumin	0.680	0.422–1.096	.113
Globulin	1.639	0.999–2.692	.051
Vitamin B6*	11.684	6.419–21.267	<.001*
Hemoglobin	0.738	0.488–1.216	.233

95% CI = 95% confidence interval, HR = hazard ratio, MTRR = Maintenance time from recovery to recurrence.
* $P < .05$.

Animal studies have shown that vitamin B6 leads to elevated homocysteine levels, which increase free radical production and oxidative stress, leading to endothelial dysfunction, decreased bone blood flow, and ultimately osteoporosis.[28]

While water-soluble vitamins are relatively safe, overuse of vitamin B6 may increase the risk of ankle fractures. The possible adverse effects of high-dose vitamin B6 supplements have been suggested before. Large doses of pyridoxine, the inactive form of vitamin B6, including those found in supplements and foods, inhibit the active form of pyridoxal phosphate. High intakes are due to the use of supplements. High doses (≥500 mg/d) may increase the risk of falls because neurological symptoms, including ataxia, neuropathy, and decreased muscle tone, Mild neurological symptoms have been observed at doses of about 100 mg/d as an adverse effect in preliminary studies, High levels of vitamin B6 may accelerate bone loss by neutralizing estrogen's role in regulating steroid receptors. The inactive form of vitamin B6 is included in supplements and is also present in foods. It inhibits the active form of pyredoxal phosphate. High doses of vitamin B6 can cause unexpected adverse reactions.[11]

Homocysteinuria is a disease characterized by high plasma homocysteine, which, due to the attachment of Hcy, usually results in the distribution of bone collagen, leading to changes in bone collagen fibers and bone fragility. Several epidemiological studies have found that increased plasma homocysteine concentrations are associated with a higher incidence of osteoporotic fractures. Multiple observational studies have found that high plasma total homocysteine levels are a potential risk factor for osteoporotic fractures.[29] The underlying mechanism between plasma homocysteine levels and fracture remains unclear. Possible mechanisms include that homocysteine regulates bone tissue quality by changing the properties of collagen cross-linking, affects bone resorption by stimulating the formation and activity of osteoclasts, and induces mitochondrial dysfunction. Vitamin B plays an important role in homocysteine metabolism. The effect of vitamin B supplementation on cysteine reduction is well known. Folic acid supplementation reduced plasma homocysteine by about 25%, while vitamin B6 also had an additional homocysteine lowering effect.

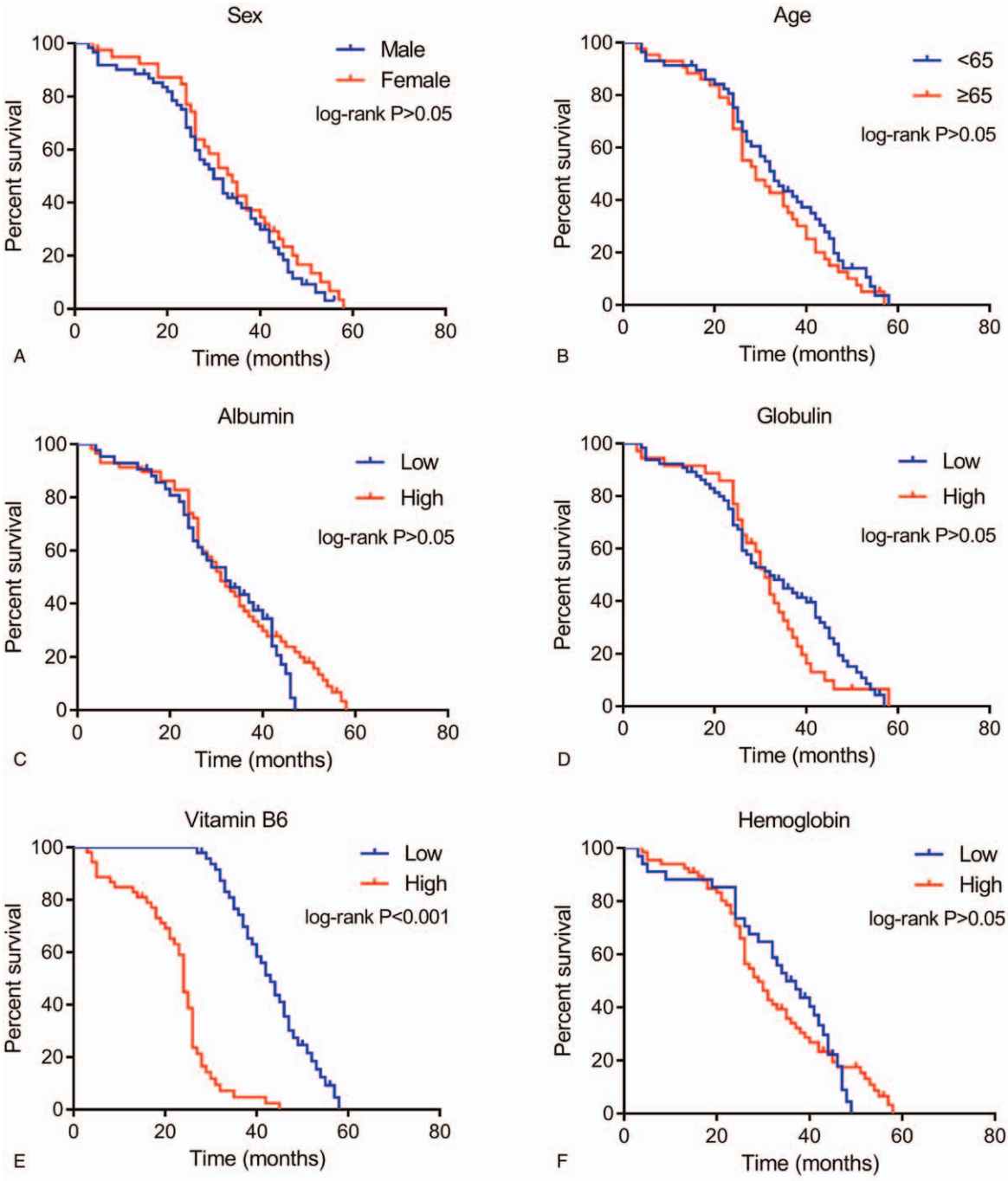

Figure 2. The overall survival by Kaplan–Meier assay. (A) sex, (B) age, (C) albumin, (D) globulin, (E) vitamin B6, (F) hemoglobin.

However, this study also has some defects. Although clinical specimens were tested and analyzed, the molecular mechanism of vitamin B6 on osteoporosis and fractured ankles was not verified in animal models. Therefore, future studies should focus on animal experiments to explore the molecular functions of vitamin B6, so as to find its molecular pathways and mechanisms in osteoporosis and fractured ankles. In addition, due to our limited funds, we did not collect indicators such as age, body mass index, etiology of osteoporosis, disease duration, treatment, etc. We will conduct further research on these indicators in order to enrich and supplement our research results.

5. Conclusions

In summary, vitamin B6 is significantly correlated with the poor prognosis of patients with osteoporosis and the increasing incidence of fractured ankles. With the increasing level of vitamin B6, patients have a poor prognosis and the increased risk of

fractured ankles might be a novel target of osteoporosis, providing new ideas for the molecular mechanism of the occurrence and development of osteoporosis.

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References

[1] Zieba JT, Chen YT, Lee BH, Bae Y. Notch signaling in skeletal development, homeostasis and pathogenesis. Biomolecules 2020;10:
[2] Malik A, Hoenig LJ. Can aging be slowed down. Clin Dermatol 2019;37:306–11.
[3] Heymann MF, Lézot F, Heymann D. The contribution of immune infiltrates and the local microenvironment in the pathogenesis of osteosarcoma. Cell Immunol 2019;343:103711.
[4] Pang KL, 0000-0003-2219-6297 AO, Chin KY, 0000-0001-6628-1552 AO. Emerging anticancer potentials of selenium on osteosarcoma. Int J Mol Sci. 2019. 20.(21).
[5] Hemminger ABK. Vitamin B6 Toxicity. StatPearls. 2020. Treasure Island. (FL).
[6] Ciapaite J, Albersen M, Savelberg SM, et al. Pyridox(am)ine 5'-phosphate oxidase (PNPO) deficiency in zebrafish results in fatal seizures and metabolic aberrations. Biochim Biophys Acta Mol Basis Dis 2020;1866:165607.
[7] Brown MMK. Vitamin B6 deficiency (Pyridoxine). StatPearls. 2020. Treasure Island.(FL).
[8] Stevelink R, Pangilinan F, Jansen FE, et al. Assessing the genetic association between vitamin B6 metabolism and genetic generalized epilepsy. Mol Genet Metab Rep 2019;21:100518.
[9] Abosamak NRV. Vitamin B6 (Pyridoxine). StatPearls. 2020. Treasure Island. (FL).
[10] Wang J, Chen L, Zhang Y, et al. Association between serum vitamin B(6) concentration and risk of osteoporosis in the middle-aged and older people in China: a cross-sectional study. BMJ Open 2019;9: e028129.
[11] Meyer HE, Willett WC, Fung TT, Holvik K, Feskanich D. Association of high intakes of vitamins B6 and B12 from food and supplements with risk of hip fracture among postmenopausal women in the nurses' health study. JAMA Netw Open 2019;2:e193591.
[12] Garcia-Ezquiaga J, Carrasco-Marina ML, Gutierrez-Cruz N, Iglesias-Escalera G, Castro-Reguera M, Perez-Gonzalez B. Pyridoxine-dependent epilepsy due to deficiency in the PNPO gene. Rev Neurol 2019;69:303–4.
[13] Mastrangelo M, Cesario S. Update on the treatment of vitamin B6 dependent epilepsies. Expert Rev Neurother 2019;19:1135–47.
[14] Yasuda H, 0000-0002-2896-7737 AO, Tsutsui M, et al. Vitamin B6 deficiency is prevalent in primary and secondary myelofibrosis patients. Int J Hematol. 2019. 110(5): 543-549.
[15] Wolke C, Gürtler S, Peter D, et al. Vitamin B6 deficiency in new born rats affects hepatic cardiolipin composition and oxidative phosphorylation. Exp Biol Med (Maywood) 2019;244:1619–28.
[16] Li J, Yin L, Wang L, et al. Effects of vitamin B6 on growth, diarrhea rate, intestinal morphology, function, and inflammatory factors expression in a high-protein diet fed to weaned piglets1. J Anim Sci 2019;97:4865–74.
[17] Dai Z, Wang R, Ang LW, Yuan JM, Koh WP. Dietary B vitamin intake and risk of hip fracture: the Singapore Chinese Health Study. Osteoporos Int 2013;24:2049–59.
[18] Hatami M, Vahid F, Esmaeil AM, et al. The vitamins involved in one-carbon metabolisms are associated with reduced risk of breast cancer in overall and subtypes. Int J Vitam Nutr Res 2020;90:131–40.
[19] Chinoy A, Mughal MZ, Padidela R. Metabolic bone disease of prematurity: causes, recognition, prevention;1; treatment and long-term consequences. Arch Dis Child Fetal Neonatal Ed 2019;104:F560–6.
[20] Yoshida T, Wang J, Stern PH. Gonadal hormones and bone. Handb Exp Pharmacol 2019.
[21] Olimpo RMC, Moretto FCF, De Sibio MT, et al. The importance of estrogen for bone protection in experimental hyperthyroidism in human osteoblasts. Life Sci 2019;231:116556.
[22] Paul RG, 0000-0002-1579-3870 AO, Hennebry AS, Elston MS, Conaglen JV, McMahon CD. Regulation of murine skeletal muscle growth by STAT5B is age- and sex-specific. Skelet Muscle. 2019. 9(1): 19.
[23] Ikeda K, Horie-Inoue K, Inoue S. Functions of estrogen and estrogen receptor signaling on skeletal muscle. J Steroid Biochem Mol Biol 2019;191:105375.
[24] Kushwaha P, Ahmad N, Dhar YV, et al. Estrogen receptor activation in response to Azadirachtin A stimulates osteoblast differentiation and bone formation in mice. J Cell Physiol 2019;234:23719–35.
[25] Saoji R, Desai M, Das RS, Das TK, Khatkhatay MI. Estrogen receptor α and β gene polymorphism in relation to bone mineral density and lipid profile in Northeast Indian women. Gene 2019;710:202–9.
[26] Xie BP, Shi LY, Li JP, et al. Oleanolic acid inhibits RANKL-induced osteoclastogenesis via ER alpha/miR-503/RANK signaling pathway in RAW264.7 cells. Biomed Pharmacother 2019;117:109045.
[27] Stone KL, Lui LY, Christen WG, et al. Effect of combination folic acid, Vitamin B(6), and Vitamin B(12) supplementation on fracture risk in women: a randomized, controlled trial. J Bone Miner Res 2017;32:2331–8.
[28] Fratoni V, Brandi ML. B vitamins, homocysteine and bone health. Nutrients 2015;7:2176–92.
[29] Ruan J, Gong X, Kong J, Wang H, Zheng X, Chen T. Effect of B vitamin (folate, B6, and B12) supplementation on osteoporotic fracture and bone turnover markers: a meta-analysis. Med Sci Monit 2015;21:875–81.