Progress in Nutrition 2022; Vol. 24, N. 4: e2022134 DOI: 10.23751/pn.v24i4.13033

ORIGINAL ARTICLE

Effect of short-term thiamine supplementation on oxidative stress, inflammation, exercise capacity and prognosis in chronic heart failure: a randomized clinical trial

Like Geng[1], Sanjun He[2], Luzhao Wang[1], Qun Dang[1], Fang Wang[1], Guo Lv[1]

[1]Department of Cardiology, Hanzhong Central Hospital, Hanzhong City, Shaanxi Province, China; [2]Department of Laboratory medicine, Hanzhong Central Hospital, Hanzhong City, Shaanxi Province, China

Abstract. *Background and aim:* Thiamine has known antioxidative and anti-inflammatory effects. However, the effectiveness of thiamine supplementation and clinical outcome in chronic heart failure (CHF) are unclear. Therefore, this study focuses on evaluating the effect of short-term thiamine supplementation on oxidative stress, inflammation, exercise capacity, and predicts the ability of rehospitalization within 30-day in patients with CHF. *Methods:* Sixty hospitalized patients with CHF were randomly divided into two groups. Both groups received conventional anti-heart failure treatment, but the thiamine group (n=30) received thiamine (100 mg/day) by intramuscular injection for 1 week, while the control group (n=30) did not do it. Serum thiamine, malondialdehyde (MDA), superoxide dismutase (SOD), high-sensitivity C-reactive protein (hs-CRP), interleukin-6 (IL-6), pro-B-type natriuretic peptide (pro-BNP) levels and 6-minute walking distance (6MWD) were detected in the two groups before and after the treatment, and all the participants were followed-up for 30-day after discharge. *Results:* After 1 week of treatment, serum thiamine levels were significantly decreased in control group compared with baseline ($p<0.05$), and the SOD levels and 6MWD in the thiamine group were significantly increased compared with the control group ($p<0.05$). Serum thiamine levels in the thiamine group were independent determinants of serum SOD levels (standardized coefficient=0.4, p=0.022) and 6MWD (standardized coefficient=0.518, p=0.004) after thiamine supplementation. Serum thiamine levels before discharge (hazard ratio [HR], 0.957, 95% confidence interval [CI], 0.924 to 0.992; p=0.016) was independently related to the rehospitalization within 30-day. *Conclusions*: In conclusion, short-term thiamine supplementation could improve oxidative stress and exercise capacity, and serum thiamine levels before discharge was an independent predictor of rehospitalization within 30-day. Meanwhile, furosemide could reduce serum thiamine levels in patients with CHF.

Key words: Thiamine; Chronic heart failure; Oxidative stress; Inflammation; Exercise capacity; Rehospitalization.

Introduction

CHF is a progressive clinical syndrome. Over the past 70 years, there have been significant advances in drugs and device therapy, survival rates for patients with CHF have improved modestly, but the 5-year mortality rate is still about 50% (1). Oxidative stress and inflammatory cytokines lead to ventricular remodeling and aggravate heart failure (2). The alleviation of inflammation and the enhancement of

antioxidant defense system contribute to the improvement heart function. Therefore, antioxidants and anti-inflammatory drugs may be potential therapies for patients with CHF.

Thiamine, or vitamin B1, is an important co-factor of metabolic enzymes, and its deficiency can lead to cardiovascular dysfunction (3). Thiamine deficiency is prevalent in patients with CHF, studies have shown that the rate of thiamine deficiency is 3% to 91% in patients with CHF (4). It is shown that the main cause of thiamine deficiency in patients with CHF is the diuretics to accelerate the excretion of thiamine in the urine (5), but there are controversies (6). It is demonstrated that thiamine deficiency can cause heart failure or aggravate the original heart failure, and deterioration of heart function will aggravate thiamine deficiency and promote oxidative stress and inflammation, thereby forming a vicious circle (7) . Therefore, it is very significant to understand thiamine levels in patients with CHF and to give appropriate thiamine supplementation.

Animal experiments have shown that thiamine deficiency may increase the production of reactive oxygen species (ROS) in rats with heart failure, and thiamine deficiency rats have increased oxidative stress and decreased antioxidant capacity (8). ROS production, free radical formation and lipid peroxidation are enhanced in thiamine deficiency, and thiamine has potential free radical cleaning activity (9). Excessive ROS may act as signaling molecules to trigger the production of pro-inflammatory cytokines in CHF (10), and thiamine deficiency is associated with increased gene expression of pro-inflammatory cytokines (11). Many studies have shown that thiamine deficiency is an important factor leading to inflammation and oxidative stress in neurodegenerative diseases, and thiamine supplementation has potential antioxidant and anti-inflammatory effects in neurodegenerative diseases (12,13). However, studies on thiamine supplementation on oxidative stress, inflammation, and prognosis in patients with CHF are rare.

The primary purpose of this study was to evaluate the effects of thiamine supplementation on oxidative stress, inflammation, and exercise capacity, as well as its ability to predict rehospitalization within 30-day in patients with CHF. In addition, we also evaluated the effect of furosemide on serum thiamine levels.

Materials and methods

Study population

This study was a randomized controlled trial that was registered at http://www.chictr.org.cn (registration number: ChiCTR1800018226). A total of 60 patients diagnosed with CHF who were hospitalized and received furosemide treatment in the Department of Cardiology, from September 2018 to October 2019 were selected and divided into thiamine group and control group using a random number table. The inclusion criteria were as follows: aged 18 to 80 years, pro-BNP>300pg/mL, left ventricular ejection fraction<40% (by echocardiography), New York Heart Association functional class □ to □, shortness of breath or orthopnea, ankle edema, interstitial transudation on chest radiography. Exclusion criteria were as follows: a major renal or liver failure, long-term drinking, pregnancy, cancer, infectious diseases, patients who cannot walk independently, an acute myocardial infarction or a cardiac surgery within the past 3 months, intake of thiamine or antioxidant supplements within the last 1 month.

All participants signed informed consent after being told the details of the study. The study complies with the Declaration of Helsinki, and the protocol was approved by the Institutional Ethics Committee (Ethical number: IRB2018-U).

Study protocol

The study was a single-blind trial. In other words, the researchers knew the drug use of the subjects, but the subjects didn't. All participants were treated with optimal anti-heart failure therapy, including daily intravenous furosemide 40 mg, and oral beta blockers, spironolactone, and benazepril. The patients in the thiamine group were received 100 mg intramuscular injection of thiamine daily based on conventional anti-heart failure therapy, and the control group received daily conventional anti-heart failure therapy for 1 week which is the course of treatment for both groups. The participants completed a baseline visit, and physical examination and laboratory tests and echocardiography were performed in the baseline period.

At the baseline period and 1 week after treatment, all participants were measured for the following indicators: serum thiamine, SOD, MDA, hs-CRP, IL-6, pro-BNP levels and 6MWD.

Patients were followed up by telephone after discharge to obtain patient survival from patients or relatives. The primary outcome was the rehospitalization due to exacerbation of CHF within 30-day after discharge. No patient died or lost to follow-up during the follow-up period. A flow diagram of the study is illustrated in Figure 1.

Serum biochemical analyses

Blood samples were collected by peripheral venous puncture at the baseline period and after the treatment for 1 week, immediately centrifuged at 3000 g for 15 minutes, and serum frozen at -80□ until assays were performed. Serum MDA levels were measured by the enzyme linked immunosorbent assay (ELISA) using Human MDA ELISA Kit (Shanghai XinYu Biotechnology Co., Ltd, Shanghai, China), following the manufacturer's instructions. Serum SOD levels were determined by pyrogallol autoxidation method with SOD assay kit (Zhongyuan Biotechnology Co. Ltd., Chongqing, China) on an automatic biochemical analyzer (model 7600; Hitachi, Ltd., Tokyo, Japan). Serum IL-6 levels were analyzed on a Roche Cobas 6000 analyzer (Roche Diagnostics, Basel, Switzerland) using the elecsys IL-6 kit (Roche Diagnostics, Mannheim, Germany) by electrochemiluminescence. Serum hs-CRP levels were measured by hs-CRP kit (Maccura Biotechnology Co., Ltd., Sichuan, China) with the latex particle-enhanced immunoturbidimetric assay on a Hitachi 7600 automatic analyzer (Hitachi, Tokyo, Japan). Serum pro-BNP levels were measured on a Roche Cobas 6000 analyzer (Roche Diagnostics, Basel, Switzerland) using elycsys pro-BNP □ kits (Roche Diagnostics, Mannheim, Germany) by electrochemiluminescence

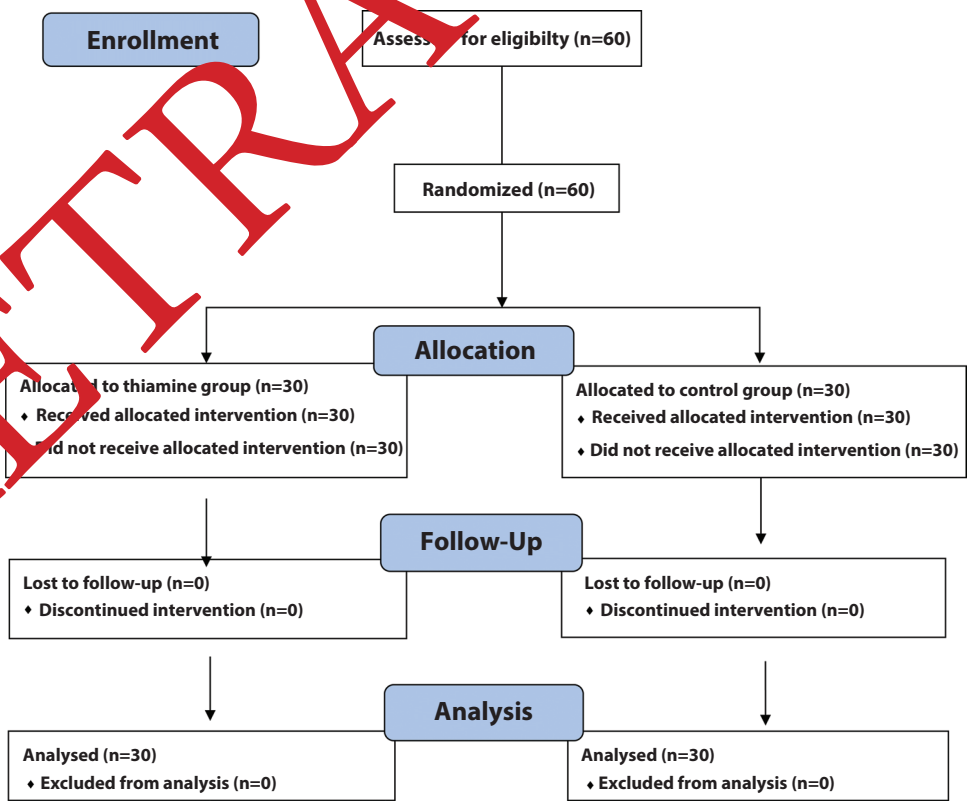

Figure 1. Participant flow during the trial.

immunoassay. Serum thiamine levels were assayed using the human Vitamin B1 ELISA kit (Shanghai XinYu Biotechnology Co., Ltd, Shanghai, China), following the manufacturer's instructions. Measure the absorbance with a Rayto-6100 microplate reader (Rayto Life and Analytical Sciences Co., Ltd., Shenzhen, China) at a wavelength of 450 nm, and the serum thiamine levels was then determined by comparing the optical density of the samples to the standard curve. The normal range of serum thiamine levels was 16 to 48 ng/mL (14). Therefore, thiamine deficiency was defined as the serum thiamine levels of less than 16 ng/mL.

Six-min walk test

The 6-minute walk test was conducted following the international standard protocol. At the baseline period, all participants were trained. Participants are instructed to walk as far as possible in 6 minutes in 70 meters obstacle-free corridor. At the baseline period and the 8th day, the trained physician tested and recorded the 6MWD.

Statistical analysis

Statistical analyses were done with SPSS for Windows, version 22.0 (SPSS Inc., USA). Normality of distribution was assessed by the Shapiro-Wilk test and by visual inspection of plotted QQ plots. Continuous data are presented as mean ± standard deviation, student's t-test was used to compare different parameters between the two groups, and paired t-test was used to compare different parameters before and after treatment in the same group. Categorical data was expressed as frequency and percentage, and Chi-square test and Fisher exact probability were used to compare categorical variables between two groups. Pearson's linear correlations were used to evaluate the correlation between serum thiamine levels and the other variables before and after treatment. Univariate and multivariate linear regression analysis were used to analyze the relationship between the serum SOD levels and 6MWD and age and serum thiamine and pro-BNP levels in the thiamine group after treatment. The rehospitalization survival curves were plotted by using the Kaplan-Meier method, and log-rank test was used to compare the difference in rehospitalization within 30-day between the two groups. Univariate and multivariate Cox proportional risk analysis were conducted to explore the risk factors of rehospitalization within 30-day. The examined variables included age and before discharge laboratory findings. The variables which showed significant correlation (P<0.1) in univariate were included in the multivariate model. Probability was assessed using a two-tailed t-test. A value of P<0.05 was considered statistically significant.

Results

Sixty hospitalized CHF patients had completed this trial. The baseline period demographic data, laboratory data and clinical characteristics were summarized in Table 1, there was no significant differences in baseline characteristics between the two groups. Thiamine has few side effects at normal doses, mainly including allergic reactions, upset stomach, feeling of warmth, restlessness, sweating a lot, weakness, irritation where the shot is given. All subjects in this study had no adverse reactions after intramuscular injection of thiamine.

After 1 week of treatment, there were 4 cases of thiamine deficiency in the control group (3 cases increased from baseline period), and no thiamine deficiency in the thiamine group. As shown in Table 2, after 1 week of treatment, the serum thiamine, SOD levels and 6MWD in the thiamine group were significantly increased compared with the baseline period (p<0.05), while MDA, IL-6, hs-CRP and pro-BNP levels were significantly decreased (p<0.05). After 1 week of treatment, the serum thiamine, MDA, IL-6, hs-CRP and pro-BNP levels were significantly decreased and 6MWD was significantly increased compared with the baseline period (p<0.05), while serum SOD levels had no significant changes in the control group (p>0.05). Compared with the control group, thiamine supplementation significantly increased the serum thiamine, SOD levels and 6MWD (p<0.05), but it did not affect the serum MDA, hs-CRP, IL-6 and pro-BNP levels(p>0.05).

Table 1. Baseline demographic, clinical characteristics and laboratory data.

Parameters	All patients (n=60)	Thiamine group (n=30)	Control group (n=30)	p
Demographics				
Number (Male/Female)	60 (40/20)	30 (19/11)	30 (21/9)	
Age (Years)	65.42±10.03	65.8±9.96	65.03±10.25	0.77
Physical features				
SBP (mmHg)	124.75±19.57	122.73±17.1	126.77±21.8	0.43
DBP (mmHg)	78.93±16.9	77.0±16.49	0.87±17.35	0.38
BMI (kg/m^2)	21.29±2.44	21.04±1.95	2.54±2.87	0.429
Echocardiography				
LVEF (%)	29.45±5.81	29.97±5.71	28.93±5.	0.496
Laboratory data				
Creatinine (mmol/L)	83.33±20.54	83.86±20.45	82.8±20.96	0.843
ALT (U/L)	34.35±15.77	31.94±14	36.76±17.15	0.24
AST (U/L)	33.0±11.86	30.07±11.0	35.91±12.07	0.056
Thiamine deficiency, (n%)	4 (6.67%)	3 (10%)	1 (3.33%)	0.612
Heart failure cause				
Ischemic cardiomyopathy	27 (45%)	13 (43.33%)	14 (46.67%)	0.795
Dilated cardiomyopathy	12 (20%)	7 (23.33%)	5 (16.67%)	0.519
Heart valve disease	14 (23.33%)	8 (26.67%)	6 (20%)	0.542
Hypertensive heart disease	6 (10%)	2 (6.67%)	4 (13.33%)	0.671
Pulmonary heart disease	1 (1.67%)	0 (0%)	1 (3.33%)	1
NYHA class				
Class III, n (%)	35 (58.33%)	16 (53.33%)	19 (63.33%)	0.432
Class IV, n (%)	25 (41.67%)	14 (46.67%)	11 (36.67%)	0.432
Medication[a]				
Beta-blocker, n (%)	22 (36.67%)	11 (36.67%)	11 (36.67%)	1
ACEI or ARB, n (%)	25 (41.67%)	13 (43.33%)	12 (40%)	0.793
Spironolactone, n (%)	28 (46.67%)	15 (50%)	13 (43.33%)	0.605
Loop diuretic, n (%)	31 (51.67%)	17 (56.67%)	14 (46.67%)	0.438

Abbreviations: Mean values ± standard deviation (SD) or percentage of patients. ALT, alanine aminotransferase; AST, aspartate aminotransferase; ACEI, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blocker. BMI, body mass index; DBP, diastolic blood pressure; FBG, fasting blood glucose; LVEF, left ventricular ejection fraction; NYHA, New York Heart Association; SBP, systolic blood pressure.
[a] Medication used in patients with chronic heart failure in the two weeks prior to hospitalization.

As shown in Table 3, according to Pearson's correlation analysis, there was no significant correlation between serum thiamine levels and serum SOD, MDA, IL-6, hs-CRP, pro-BNP levels and 6MWD in both groups at the baseline period. After 1 week of treatment, the serum thiamine levels in the thiamine group were significantly positively correlated with serum SOD levels (r=0.448, p=0.013) and 6MWD (r=0.596, p=0.001), and negatively correlated with serum pro-BNP levels (r =-0.39, p=0.033). However, the serum thiamine level in the control group was not correlated with all indicators.

As shown in Table 4, simple linear regression analysis showed that serum SOD levels were significantly positively correlated with serum thiamine levels (unstandardized coefficient=0.479, p=0.013),

Table 2. Serum thiamine levels and various indicators in the two groups at baseline and 1 week after thiamine supplementation.

Variable	Thiamine Group		Control Group	
	Baseline	1 week	Baseline	1 week
Thiamine (ng/mL)	29.16±7.83	75.19±16.18§	29.26±5.2	23.92±7.95□★
SOD (U/mL)	102.1±12.68	115.26±17.31§	101.02±14.85	104.85±12.81★
MDA (nmol/mL)	4.87±0.76	4.22±0.56§	4.8±0.84	4.46±0.6□
IL-6 (pg/mL)	11.36±3.33	6.75±2.21§	11.29±3.2	7.61±2.33□
hs-CRP (mg/L)	9.25±2.89	5.36±1.6§	9.56±2.4	97±1.31□
pro-BNP (pg/mL)	12340.7±6597.8	893.1±247.2§	114[illegible]±6819.7	892.8±197□
6MWD (m)	193.3±22.6	410.7±19.7§	196.5±25.7	391.4±37.9□★

Abbreviations: Values are means ± standard deviation (SD). hs-CRP, high-sensitivity C-reactive protein; IL-6, Interleukin-6; MDA, malondialdehyde; pro-BNP, pro-B-type natriuretic peptide; SOD, superoxide dismutase; 6MWD, 6-minute walking distance.
§P<0.05, versus baseline period in thiamine group.
□P<0.05, versus baseline period in control group.
★P<0.05, versus after thiamine supplementation in the thiamine group.

Table 3. Correlation between serum thiamine levels and various indicators in the two groups before and after treatment.

Variable	Thiamine Group Baseline		Thiamine Group 1 week		Control Group Baseline		Control Group 1 week	
	r	p	r	p	r	p	r	p
SOD (U/mL)	0.195	0.302	0.448	0.013	0.25	0.183	0.323	0.082
MDA (nmol/mL)	-0.186	0.325	-0.3	0.108	-0.025	0.894	-0.231	0.219
IL-6 (pg/mL)	-0.068	0.771	-0.159	0.401	-0.335	0.07	-0.265	0.157
hs-CRP (mg/L)	0.118	0.533	-0.251	0.181	-0.072	0.704	0.013	0.947
pro-BNP (pg/mL)	-0.324	0.081	-0.39	0.033	-0.263	0.16	-0.314	0.091
6MWD (m)	0.311	0.093	0.596	0.001	0.116	0.542	0.29	0.12

Abbreviations: hs-CRP, high-sensitivity C-reactive protein; IL-6, Interleukin-6; MDA, malondialdehyde; pro-BNP, pro-B-type natriuretic peptide; SOD, superoxide dismutase; 6MWD, 6-minute walking distance.

Table 4. Univariable and multivariate regression analysis of serum SOD levels and 6MWD in thiamine group after thiamine supplementation for 1 week.

Variable	SOD				6MWD			
	Univariable		Multivariable		Univariable		Multivariable	
	B	P	β	P	B	P	β	P
Age (Years)	0.624	0.051	0.294	0.086	0.281	0.453	—	—
pro-BNP (pg/mL)	0.013	0.333	—	—	-0.032	0.027	-0.201	0.23
Thiamine (ng/mL)	0.479	0.013	0.40	0.022	0.725	0.001	0.518	0.004
			Adjusted R^2=0.232				Adjusted R^2=0.344	

Abbreviations: B, unstandardized coefficient; β, standardized coefficient; pro-BNP, pro-B-type natriuretic peptide; SOD, superoxide dismutase; 6MWD, 6-minute walking distance.

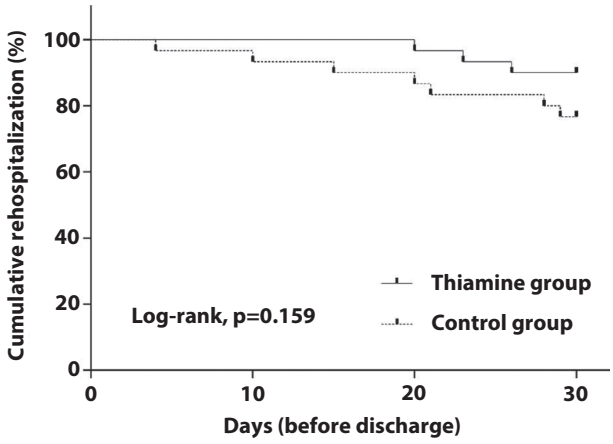

Figure 2. Kaplan-Meier curve of 30-day rehospitalization rate for both groups.

while 6MWD were significantly negatively correlated with serum pro-BNP levels (unstandardized coefficient=-0.032, p=0.027) and significantly positively correlated with serum thiamine levels (unstandardized coefficient=0.725, p=0.001). Further, a multivariate linear regression analysis showed that the serum thiamine levels had a significant effect on SOD (standardized coefficient=0.4, p=0.022) and 6MWD (standardized coefficient=0.518, p=0.004) in the thiamine group after 1 week of treatment.

During the 30-day follow-up period, a total of 10 patients in both groups were rehospitalization for CHF deterioration (3 in the thiamine group and 7 in the control group). As shown in Figure 2, although the thiamine group seemed to have advantages, Kaplan-Meier curve analysis showed that rehospitalization rate for heart failure exacerbation no significant difference between the two groups after follow-up of 30-day (log-rank, p=0.159).

As shown in Table 5, in univariate Cox regression analysis, serum thiamine levels (HR, 0.955; 95% CI, 0.918 to 0.993; p=0.021) before discharge (1 week after treatment) and pro-BNP levels (HR, 1.003; 95% CI, 1.0 to 1.006; p=0.048) were associated with rehospitalization within 30-day. Further, a multivariate Cox regression analysis showed that serum thiamine levels before discharge (HR, 0.957; 95% CI, 0.924 to 0.992; p=0.016) was independently associated with rehospitalization within 30-day, but pro-BNP did not.

Discussion

Our study mainly demonstrated that serum thiamine levels was a significant influence factor of SOD and 6MWD. In addition, serum thiamine levels before discharge was independently associated with rehospitalization within 30-day due to exacerbation of heart failure. Last, furosemide increased serum thiamine levels.

The active form of thiamine in the body is thiamine diphosphate (ThDP), it acts as a coenzyme for transketolase (TKT) and for the α-ketoglutarate dehydrogenase and pyruvate dehydrogenase complex, enzymes that are key rate-limiting enzyme in the Krebs cycle and play a basic role in myocardial energy metabolism (15). ROS mediates cardiac remodeling, and elevated ROS is the main source of oxidative stress (16), while nicotinamide adenine dinucleotide phosphate (NADPH) oxidase has been shown to be the main source of ROS production in heart failure (17). TKT is a ThDP (thiamine) dependent enzyme. Studies have shown that TKT activity is significantly reduced when thiamine is deficient (18), and the activity of TKT is increased after thiamine supplementation (19). TKT promotes the conversion from glycolysis to the pentose phosphate pathway, resulting in a reduction in NADPH oxidase and production in ROS (20). Studies had been shown that benfotiamine, a derivative of thiamine, could reduce oxidative stress in mice with myocardial infarction by inhibiting the action of NADPH oxidase (21). Therefore, in this study, thiamine supplementation increased serum ThDP concentration, resulting in enhancing TKT activity and reducing NADPH oxidase and ROS production, thus reducing oxidative stress levels in patients with CHF. SOD and MDA are the main markers of oxidative stress used to evaluate the ability to eliminate oxygen free radicals. Sarandol E et al treated diabetic rats with thiamine (6 mg/kg) in drinking water for 5 weeks, and the results showed that the level of MDA in plasma and tissues in the thiamine group was significantly lower than that in the control group, and the activity of SOD was significantly increased (22). Radonjic T et al. injected thiamine hydrochloride (25 mg/kg) into the abdominal cavity of doxorubicin-treated rats for 7 days, they found that the SOD activity of rats

Table 5. Predict the 30-day rehospitalization rate by univariate and multivariate Cox regression analysis.

Variable	Univariable		Multivariable	
	HR (95% CI)	p	HR (95% CI)	p
Age (Years)	1.001 (0.94 to 1.065)	0.983	—	—
Thiamine (ng/mL)	0.955 (0.918 to 0.993)	0.021	0.957 (0.924 to 0.992)	0.016
SOD (U/mL)	0.969 (0.93 to 1.01)	0.139		—
MDA (nmol/mL)	1.844 (0.618 to 5.497)	0.272	—	—
IL-6 (pg/mL)	1.097 (0.84 to 1.433)	0.497	—	—
hs-CRP (mg/L)	0.922 (0.605 to 1.405)	0.706		—
pro-BNP (pg/mL)	1.003 (1.0 to 1.006)	0.048	1.003 (1.0 to 1.007)	0.053
6MWD (m)	0.992 (0.973 to 1.01)	0.373	—	—

Abbreviations: hs-CRP, high-sensitivity C-reactive protein; IL-6, Interleukin-6; MDA, malondialdehyde; pro-BNP, pro-B-type natriuretic peptide; SOD, superoxide dismutase; 6MWD, 6-minute walking distance.

was significantly rose, while the antioxidant defense system was increased, and the myocardial contractility was enhanced (23). Similar to the results of those reports, our study has found that the serum SOD levels of thiamine group were significantly increased than the control group after 1 week, but MDA levels were no significant difference. The main reason may be our research intervention time was short, changes in MDA may not have been obvious and had sufficient time to fully develop.

Gonzalez-ortiz M et al conducted a randomized, placebo-controlled clinical trial in 24 patients with type 2 diabetes mellitus, in which 12 subjects received thiamine orally once daily (100 mg) and 12 patients (control group) received placebo for 1 month which was no significant difference in IL-6 and hs-CRP (24). This study was consistent with our findings that thiamine supplementation did not significantly affect serum hs-CRP and IL-6 levels in patients with CHF. However, Amirani E et al divided 60 patients with gestational diabetes into two group, the intervention group was received thiamine supplementation (100 mg/day) and the control group was received placebo for 6 weeks, and they found that plasma hs-CRP levels and TNF-α gene expression were significantly reduced in the thiamine group (25). The difference between the results of such trials may be caused for differences in study design, different durations of trials, different doses of thiamine used, and specific characteristics of participants.

Smithline HA et al injected thiamine (100 mg/day) for 2 days in patients with acute heart failure without thiamine deficiency, and the results showed that there was no significant decrease in pro-BNP in the thiamine group compared with the control group (26). Keith M et al randomly divided 69 cases of CHF patients with reduced ejection fraction into two groups, 35 cases of thiamine group were given oral thiamine 200 mg daily, 34 cases of control group were given placebo, after 6 months of treatment, there were no significant difference in 6-MWD and pro-BNP between the two groups(27). Our study found that short-term thiamine supplementation significantly improved 6MWD in patients with CHF, but it did not significantly improve the serum pro-BNP levels. There are many possible explanations for this result. First, thiamine is the essential factor for energy production, especially in the heart, and thiamine supplementation increases myocardial adenosine triphosphate levels (28), thereby improving exercise capacity in patients with CHF. Secondly, in our study, the serum thiamine levels of the control group were significantly reduced compared with the baseline period after 1 week of furosemide treatment, but it did not reach the level of thiamine deficiency. The deterioration of cardiac function mediated by thiamine deficiency did not occur, so there was no significant change in pro-BNP levels.

The study also assessed the effects of pre-discharge indicators on 30-day rehospitalization for CHF because of deterioration of heart failure. There was no

significant difference in 30-day rehospitalization rates between the two groups, but serum thiamine levels before discharge were related with 30-day rehospitalization in patients with CHF. The relationship between serum thiamine levels and prognosis in patients with CHF has not been reported. However, studies have shown that early thiamine use can improve short-term survival in critically ill patients with acute kidney injury (29). Our study has found the similar results in patients with CHF. Studies have shown that thiamine insufficiency, which is milder than thiamine deficiency, does not cause classic clinical symptoms, but it increases the risk of CHF (30).

Our findings were that there were 4 cases (6.67%) of thiamine deficiency at the baseline period in the two groups. Although we expected that many patients with thiamine deficiency would appear in the control group after 1 week of furosemide treatment, the result was that the 30 participants in the control group only have 3 new patients with thiamine deficiency after that. Although thiamine levels in the control group did not reach the level of deficiency, they were significantly lower than at the baseline period, it was indicated that furosemide promoted thiamine excretion. This result was similar to the reported by Seligmann H et al. their study showed that furosemide treatment resulted in a significant reduction in thiamine level (31). However, Yue QY et al showed that there was no difference in serum thiamine levels between heart failure patients who received furosemide therapy and patients without heart failure, and thiamine deficiency may not be caused by furosemide therapy (6). Differences between the results of the various trials may be related to differences duration of the trials, different doses of diuretics used, sample size, and average daily dietary thiamine intake. Therefore, the relationship between diuretics and thiamine levels in patients with CHF needs further study.

This study has several limitations. First, thiamine cannot be synthesized by humans, and thiamine is mainly consumed through diet, body stores are limited (32). However, during the study period, we did not adopt a uniform diet for the participants, so the dietary thiamine intake of each participant was different. Second, in this study, the duration of thiamine intervention was 7 days, and short-term thiamine supplementation may not be enough to significantly influenced on MDA, hs-CRP, IL-6 and pro-BNP. Third, the sample size of our study is limited, which reduces the statistical power of COX regression analysis. Fourth, although all participants were followed up for 30-day, all participants did not continue to take thiamine after discharge. Fifth, this study adopted blank control, which may lead to certain bias in the results. Sixth, in this study, serum thiamine levels were measured by ELISA, but studies showed that thiamine levels in foods and medicines measured by ELISA and liquid chromatography coupled to tandem mass spectrometry which allows sensitivity and accurate measurement serum thiamine were highly correlated (33). Therefore, ELISA is a reliable and accurate method to detect serum thiamine levels. In a word, long duration, placebo-controlled, double-blind, large sample size, randomized controlled trial are needed to further study.

Conclusion

In conclusion, the most important finding is that thiamine supplementation could improve oxidative stress and exercise capacity in hospitalized patients with CHF in the short-term, but could not improve inflammation. Second, serum thiamine levels before discharge were independently associated with rehospitalization within 30-day due to exacerbation of heart failure. In addition, furosemide could significantly reduce serum thiamine levels. Therefore, thiamine supplementation is beneficial for patients with symptomatic CHF treated with diuretics, and we suggest that thiamine could be used as an adjuvant therapy for CHF.

Acknowledgements: We thank the participants in the study for their cooperation.

Conflict of Interest: Each author declares that he or she has no commercial associations (e.g. consultancies, stock ownership, equity interest, patent/licensing arrangement etc.) that might pose a conflict of interest in connection with the submitted article.

References

1. Jones NR, Roalfe AK, Adoki I, Hobbs FDR, Taylor CJ. Survival of patients with chronic heart failure in the community: a systematic review and meta-analysis. Eur J Heart Fail. 2019; 21: 1306-25.
2. Aimo A, Castiglione V, Borrelli C, et al. Oxidative stress and inflammation in the evolution of heart failure: From pathophysiology to therapeutic strategies. Eur J Prev Cardiol. 2020; 27: 494-510.
3. Eshak ES, Arafa AE. Thiamine deficiency and cardiovascular disorders. Nutr Metab Cardiovasc Dis. 2018; 28: 965-72.
4. Goel A, Kattoor AJ, Mehta JL. Thiamin therapy for chronic heart failure: is there any future for this vitamin? Am J Clin Nutr. 2019; 110: 1270-71.
5. Katta N, Balla S, Alpert MA. Does Long-Term Furosemide Therapy Cause Thiamine Deficiency in Patients with Heart Failure? A Focused Review. Am J Med. 2016; 129: 753 e7-53 e11.
6. Yue QY BB, Lindström B, Nyquist O. No difference in blood thiamine diphosphate levels between Swedish Caucasian patients with congestive heart failure treated with furosemide and patients without heart failure. J Intern Med. 1997; 242: 491-95.
7. Hanninen SA, Darling PB, Sole MJ, Barr A, Keith ME. The prevalence of thiamin deficiency in hospitalized patients with congestive heart failure. J Am Coll Cardiol. 2006; 47: 354-61.
8. Gioda CR, de Oliveira Barreto T, Primo [illegible] Cardiac oxidative stress is involved in heart failure induced by thiamine deprivation in rats. Am J Physiol Heart Circ Physiol. 2010; 298: H2039-47.
9. Okai Y H-OK, Sato EF, Kobra R, Inoue M. Potent radical-scavenging activities of thiamin and thiamin diphosphate. J Clin Biochem Nutr 2007; 40: 42-8.
10. Ayoub KF, Pothineni NVK, Rutland J, Ding Z, Mehta JL. Immunity, Inflammation, and Oxidative Stress in Heart Failure: Emerging Molecular Targets. Cardiovascular Drugs and Ther. 2017; 31: 593-608.
11. Kruppa AJ, Suender SS, Shi Q, Xu H, Gibson G E. Changes in [illegible] processes associated with selective vulnerability following mild impairment of oxidative metabolism. Neurobiol Dis. 2007; 26: 353-62.
12. Liu D, Ke Z, Luo J. Thiamine Deficiency and Neurodegeneration: the Interplay Among Oxidative Stress, Endoplasmic Reticulum Stress, and Autophagy. Mol Neurobiol. 2017; 54: 5440-48.
13. Abdou E, Hazell AS. Thiamine deficiency: an update of pathophysiologic mechanisms and future therapeutic considerations. Neurochem Res. 2015; 40: 353-61.
14. Lynch PL, Young IS. Determination of thiamine by high-performance liquid. J Chromatogr A. 2000; 881: 267-84.
15. Manzetti S, Zhang J, van der Spoel D. Thiamin function, metabolism, uptake, and transport. Biochemistry. 2014; 53: 821-35.
16. Dey S, DeMazumder D, Sidor A, Brian Foster D, O'Rourke B. Mitochondrial ROS Drive Sudden Cardiac Death and Chronic Proteome Remodeling in Heart Failure. Circ Res. 2018; 123: 356-71.
17. Li JM, Gall NP, Grieve DJ, Chen M, Shah AM. Activation of NADPH oxidase during progression of cardiac hypertrophy to failure. Hypertension. 2002; 40: 477-84.
18. Zhao Y, Pan X, Zhao J, Wang Y, Peng Y, Zhong C. Decreased transketolase activity contributes to impaired hippocampal neurogenesis induced by thiamine deficiency. J Neurochem. 2009; 111: 537-46.
19. Alam SS, Riaz S, Waheed A, Akhtar M. Effect of high dose thiamine therapy on activity and molecular aspects of transketolase in Type 2 diabetic patients. African Journal of Biotechnology. 2011; 10:17[illegible]-16.
20. Riyaphan D, Pongchai D, Muangsombut V, et al. Transketolase and vitamin B1 influence on ROS-dependent neutrophil extracellular traps (NETs) formation. PLoS One. 2019; 14: 1-17.
21. Ahmed LA, Hassan OF, Galal O, Mansour DF, El-Khatib A. Beneficial effects of benfotiamine, a NADPH oxidase inhibitor, in isoproterenol-induced myocardial infarction in rats. PLoS One. 2020; 15:1-17.
22. Sarandol E, Tas S, Serdar Z, Dirican M. Effects of thiamine treatment on oxidative stress in experimental diabetes. Bratisl Lek Listy. 2020; 121: 235-41.
23. Radonjic T, Rankovic M, Ravic M, et al. The Effects of Thiamine Hydrochloride on Cardiac Function, Redox Status and Morphometric Alterations in Doxorubicin-Treated Rats. Cardiovasc Toxicol. 2020; 20: 111-20.
24. González-Ortiz M, Martínez-Abundis E, Robles-Cervantes JA, Ramírez-Ramírez V, Ramos-Zavala MG. Effect of thiamine administration on metabolic profile, cytokines and inflammatory markers in drug-naive patients with type 2 diabetes. Eur J Nutr. 2011; 50: 145-9.
25. Amirani E, Aghadavod E, Shafabakhsh R, et al. Anti-inflammatory and antioxidative effects of thiamin supplements in patients with gestational diabetes mellitus. J Matern Fetal Neonatal Med. 2020: 1-6.
26. Smithline HA, Donnino M, Blank FSJ, et al. Supplemental thiamine for the treatment of acute heart failure syndrome: a randomized controlled trial. BMC Complementary and Alternative Medicine. 2019;19:1-11.
27. Keith M, Quach S, Ahmed M, et al. Thiamin supplementation does not improve left ventricular ejection fraction in ambulatory heart failure patients: a randomized controlled trial. Am J Clin Nutr. 2019; 110: 1287-95.
28. Yamada Y KY, Akaoka M, Watanabe M, et al. Thiamine treatment preserves cardiac function against ischemia injury via maintaining mitochondrial size and ATP levels. J Appl Physiol 2021; 130: 26-35.
29. Li X, Luan H, Zhang H, et al. Associations between early thiamine administration and clinical outcomes in critically ill patients with acute kidney injury. The British journal of nutrition. 2021: 1-9.

30. Ao M YK, Ohta J, Abe Y, et al. Possible involvement of thiamine insufficiency in heart failure in the institutionalized elderly. J Clin Biochem Nutr. 2019; 64: 239-42.
31. Seligmann H, Halkin H, Rauchfleisch S, et al. Thiamine deficiency in patients with congestive heart failure receiving long-term furosemide therapy: A pilot study. Am J Med. 1991; 91: 151-55.
32. Whitfield KC, Bourassa MW, Adamolekun B, et al. Thiamine deficiency disorders: diagnosis, prevalence, and a roadmap for global control programs. Ann N Y Acad Sci. 2018; 1430: 3-43.
33. Zeng L, Wu X, Liu L, Xu L, Kuang H, Xu C. Production of a monoclonal antibody for the detection of vitamin B1 and its use in an indirect enzyme-linked immunosorbent assay and immunochromatographic strip. J Mater Chem B. 2020; 8: 1935-43.

Correspondence:
Sanjun He, PhD
Department of Laboratory medicine, Hanzhong Central Hospital, No. 557, Labor West Road, Hanzhong City, Shaanxi Province, 723000, China.
Phone: +8617392[illegible]1599;
E-mail: hesanjun202[illegible]@hotmail.com