

A Pilot study on correlation between Zinc and Magnesium serum concentrations and coronary slow flow phenomenon

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Abstract. *Background:* The pathophysiology of slow flow includes microvascular disorders, endothelial dysfunction, subclinical atherosclerosis, inflammation and anatomical factors. The role of magnesium and zinc in the development of microvascular and endothelial dysfunctions as well as atherosclerosis has been proven in previous studies, and the mechanism of the development has been studied. The aim of current study was to evaluate the serum concentration of zinc and magnesium in patients with epicardial coronary artery slow flow. *Design:* 125 patients who referred to Ghaem Hospital in Mashhad were selected based on inclusion and exclusion criteria. Magnesium and Zinc levels were evaluated in patients. The plasma levels of studied elements were compared among the different groups and the rate of coronary artery slow flow was evaluated based on the TIMI score. *Results:* The results of present study indicated that the serum level of Magnesium in the studied groups did not show a significant correlation with rate of coronary artery slow flow ($P > 0.05$). Serum Zinc concentration was significantly different in the studied groups, which means serum Zinc level in patients without coronary artery occlusion and without epicardial slow flow were significantly higher than other groups ($P > 0.01$). *Conclusion:* In the present study, no significant relationship was found between the serum level of zinc and magnesium with the intensity of coronary artery slow flow based on TIMI, and further studies seem to be needed to investigate this relationship.

Key words: Magnesium - Zinc - Slow flow - coronary artery disease – trace elements

Introduction

The Coronary artery slow flow phenomenon (CSFP) is an angiographic finding that is characterized by slow-forward movement of the contrast media to the end branches of an epicardial coronary artery in the absence of occlusive coronary artery disease (1). There is currently no information available on the cause and pathophysiology of CSFP. However, it has been shown that factors such as microvascular angina, vascular endothelial dysfunction, subclinical atherosclerosis,

inflammation and anatomical factors may be important in the pathophysiology of the disease (1). Microvascular angina and vascular endothelial dysfunction role are more pronounced in this phenomenon.

Although the microvascular angina pathogenesis is not well known, probably there are two simultaneous mechanisms cause pain in patient; myocardial ischemia (due to the dysfunction of the microvascular coronary artery) and increased sensitivity to chest pains (2, 3).

The vascular endothelial disorder is an important etiology for the development of microvascular

coronary artery dysfunction in patients suffering from microvascular angina. The main cause of endothelial disorders can be attributed to an imbalance in the production and consumption of nitric oxide (NO). NO consumption increases compared to its production in patients suffering from endothelial disorders, resulting in an inability of vasodilation that develops as ischemia. In fact, increased oxidative stress and inflammation lead to altered metabolism of NO in such patients (4).

Currently, counting the TIMI frames as a quantitative index of blood flow in vascular angiography is the only diagnostic and evaluator instrument, however, the permission of the clinical follow-up and long-term assessment are not possible due to the invasive nature of this method (5).

The role of magnesium and zinc in the development of microvascular and vascular endothelial dysfunction as well as atherosclerosis has been proven in previous studies. Magnesium can prevent or delay the creation of atherosclerotic plaques (6). Moreover, it can also affect the production of nitric oxide and prostaglandins, thus protecting the vascular endothelium, especially coronary arteries (7). Zinc also affects oxidative stress and reduces the oxidation of LDL, which has a protective effect against atherosclerosis. Other mechanisms include regulation of nitric oxide levels in vascular endothelium and NF- κ B levels (8). So far, there has been no study on the role of these two elements in the phenomenon of CSFP and its occurrence. The present study aims to investigate the level of zinc and magnesium in patients with occlusive and none-occlusive coronary artery disease, as well as CSFP and non-CSFP patients.

Method

Study population

We conducted this study from July 2016 until November 2017. Based on the inclusion and exclusion criteria, the patients attending the Ghaem Hospital, affiliated to Mashhad University of Medical Sciences, Mashhad, Iran for angiography are the subjects of the present study. Patients who were suspected to have coronary artery disease were included in study. Patients with coronary artery aneurysm, hyperhomocysteinemia, myocarditis, pericarditis, cardiomyopathy and sitagliptin use history or who were on hemodialysis were excluded from the study. All of the participants signed a written informed consent before the beginning of the study.

This research was approved by the Ethics Committee of Mashhad University of Medical Sciences (code number: IR.MUMS.REC.1395.599).

Determination of zinc and magnesium serum concentration and coronary blood flow speed

The existence or absence of the coronary obstructive artery disease, number of involved arteries and coronary slow flow arteries were evaluated as the visual findings during the angiography by a cardiologist.

Different definitions for the slow blood flow of coronary arteries are available that the one is used in this study is “the corrected TIMI frame counts more than two standard deviations from the normal range in the absence of an obstruction in the coronary arteries” (2). According to the result of the angiography, the patients were divided into 5 groups as mentioned in Table 1.

Table 1. classification of study population

Group's Acronym	The group of patients
CAD(-), Slow Flow(-)	Patients with no coronary artery disease and no epicardial arteries slow flow
CAD(-), Slow Flow(+)	Patients with no coronary artery disease and with the slow flow of epicardial arteries
CAD(+), (<50%),Slow Flow(+)	Patients with coronary artery disease (less than 50%) having no slow flow of epicardial arteries
CAD(+), (>50%, <90%),Slow Flow(-)	Patients with coronary artery disease (between 50 to 90%) having no slow flow of epicardial arteries
CAD(+), Slow Flow(+)	Patients with coronary artery disease having the slow flow of epicardial arteries

Twenty milliliters of patients' whole blood was collected from brachial vein, transferred to the Ghaem hospital emergency ward laboratory and centrifuged at 3000 rpm for 10 min. The plasma fraction was isolated and stored at -80°C until required for analysis. Serum concentration of zinc (AA240FS, Varian, Australia) using the atomic absorption flame spectrophotometry and magnesium (General biochemistry, Pars Azmun, Iran) were determined by magnesium kit. Patients' demographic data including gender, age and body mass index (BMI), past medical history (e.g. hypertension, dyslipidemia, and diabetes Mellitus), smoking and family history were collected using patients' records in a pre-designed checklist.

Statistical methods

Statistical analysis was carried out by SPSS 19. Results have been shown as mean \pm standard deviation or median (interquartile range) for normally and non-normally distributed continuous variables respectively, and number (percentages) for nominal variables. Kolmogorov–Smirnov test was used to assess the normality of the variables distributions.

Independent sample t-test and Mann–Whitney U-test were used respectively to compare normally and non-normally distributed variables between the two groups. For comparison of more than two groups in normally and non-normally distributed variable one way ANOVA and Kruskal–Wallis tests were used respectively.

Besides, the Pearson correlation test (in the case of normal distribution) and Spearman's correlation test (in the case of non-normal distribution) were employed to examine the intensity and correlation between the two quantitative variables. All the tests were conducted at the significant level of 0.05.

Results

A total of 125 patients undergoing angiography were enrolled in the study. Of these, 48 (38.4%) were male the mean age of the studied population was 54.58 ± 10.23 years old. As mentioned in the Table 2, most of patients had no CAD and CSFP (30.4%),

followed by those with coronary obstructive disease less than 50% and slow flow of epicardial artery (20%).

Most of patients with coronary obstruction rate of 50–90% had one artery involvement (39.1%), followed by two vessels (34.8%) and three vessels (26.1%) respectively.

Serum magnesium level was not significantly different between the studied groups ($P = 0.89$). But, the mean serum zinc level was significantly different between these groups ($P < 0.001$) (Table 3). To determine between which groups the serum level of zinc was significantly different we use post hoc tukey test. As it is mentioned in table 4, the serum level of zinc was significantly higher in CAD and Slow flow negative groups in comparison with all other groups.

TIMI score was measured in various coronary arteries separately (LCX, LAD, and RCA) in patients with a slow flow coronary artery.

Considering that two or three vessels had a slow flow in some patients, mean TIMI values were calculated individually.

Table 5 indicates that both serum levels of magnesium ($P = 0.58$) and zinc ($P = 0.39$) had no significant relationships with the severity of the slow flow of the coronary arteries based on TIMI scores in study population (Figure 1&2).

Discussion

Previous studies have shown that magnesium and zinc serum level are important in the development of microvascular and endothelial disorders, as well as atherosclerosis. Since there is a relationship between CSFP

Table 2. Distribution of patients in the studied groups

Group	Frequency (%)	Distribution (N)
CAD(-),Slow Flow(-)	30.4	38
CAD(-),Slow Flow(+)	17.6	22
CAD (+)($<50\%$),Slow Flow(-)	20	25
CAD (+)($>50\%$, $<90\%$),Slow Flow(-)	18.4	23
CAD(+),Slow Flow(+)	13.6	17
Total	100	125

Table 3. Comparison of mean magnesium concentration and serum zinc concentration in the studied groups

Group	Serum magnesium level (µg/dl) (mean ± SD)	Serum zinc level (µg/dl) (mean ± SD)
CAD(-),Slow Flow(-)	1.0±72.19	130.18±35.77
CAD(-),Slow Flow(+)	1.0±73.35	96.33±70.75
CAD(+)(<50%),Slow Flow(-)	1.0±67.20	77.39±70.75
CAD(+)(>50%,<90%),Slow Flow(-)	1.0±68.22	83.33±91.52
CAD(+),Slow Flow(+)	1.0±71.22	89.26±91.03
P value*	0.89	0.01*

*one-way ANOVA test; CAD: Coronary Arterial Disease

Table 4. Comparison of mean serum level of zinc in the studied groups based on post hoc Tukey test

STUDY gROUP		p-value*
Cad(-),Slow Flow(-)	CAD(-),Slow Flow (+)	0.001*
	CAD(+)(<50%),Slow Flow(-)	<0.001*
	CAD(+)(>50%,<90%),Slow Flow(-)	<0.001*
	CAD(+),Slow Flow(+)	<0.001*
CAD(-),Slow Flow(+)	CAD(+)(<50%),Slow Flow(-)	0.20
	CAD(+)(>50%,<90%),Slow Flow(-)	0.61
	CAD(+),Slow Flow(+)	0.956
CAD(+)(<50%),Slow Flow(-)	CAD(+)(>50%,<90%),Slow Flow(-)	0.70
	CAD(+),Slow Flow(+)	0.97
CAD(+)(>50%,<90%),Slow Flow(-)	CAD(+),Slow Flow(+)	0.95

*Post hoc tukey test

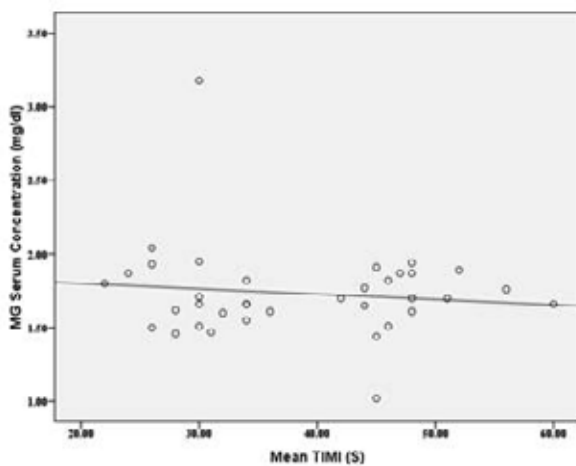


Figure 1. Correlation of serum magnesium level and TIMI score in study population

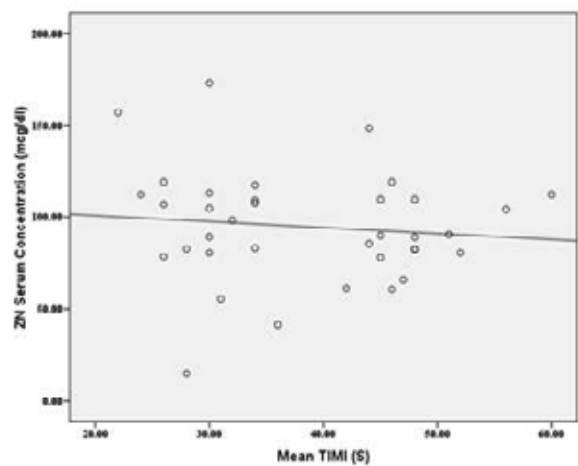


Figure 2. Correlation of serum zinc level and TIMI score in study population

Table 5. Relationship between serum zinc and magnesium concentrations with intensity of coronary artery flow based on TIMI

parameter	The correlation coefficient	p-value*
Serum magnesium Concentration	0.09	0.58
Serum zinc Concentration	-0.13	0.39

*: *Pearson correlation*

pathophysiology and the above disorder and to the best of our knowledge there is no study on the magnesium and zinc serum level correlation with CSFP, it seems necessary to investigate levels of magnesium and zinc in patients with a coronary slow flow phenomenon. It has been observed that magnesium is effective in regulation of the calcium entry into the heart and muscle cells. On the other hand, magnesium with various mechanisms such as effect on oxidative stress, LDL reuptake and metabolism and accelerated endothelial cell proliferation affects vascular endothelium performance and its low concentrations can lead to abnormal vascular endothelial activity (9,10). It also regulates the production of nitric oxide and prostaglandins, thereby affecting the endothelium of vessels, especially coronary arteries. The role of magnesium in the prevention and regulation of endothelial cell apoptosis has also been proven and seems to be one of the mechanisms of the effect of this element to prevent atherosclerosis and coronary artery disease (11,12). Several factors are presented to be involved in the pathophysiology of the CSFP, including microvascular disorders, endothelial dysfunction, subclinical atherosclerosis, inflammation and anatomical factors (13). Based on this information, it was assumed that there was a relationship between serum magnesium concentration and the CSFP, which was not seen in the present study. The reason for this may be due to the limited study population in the present study, as well as the normal serum level of magnesium in patients. The effects of magnesium on the slow flow of coronary arteries have not been studied directly in previous studies. As indicated in the results, patients were normo-magnesemia. In 1998, 13922 patients were studied for 4-7 years and the results suggested that low level of magnesium can be considered

as a predictor of coronary artery disease (14). In a systematic review study published by Delgobo et al in 2013, the serum concentration and magnesium intake in the diet and its relevance to the risk of cardiovascular disease were studied, which were associated with incidence of coronary artery disease (15).

Zinc is another trace element which is proposed to be effective on vascular endothelium function, and inflammation and atherosclerosis occurrence, as well as magnesium. So, it may contribute to the pathophysiology of the CSFP and it is assumed that there is a probable relationship between serum zinc level and the CSFP. However, we found no significant correlation between zinc level and intensity of CSFP. But it should be mentioned that the serum level of zinc was significantly higher in CAD & slow flow negative patients. So, it seems that zinc serum level may be effective in occurrence of coronary artery disease and also CSFP. In previous studies, it has been shown that in patients with coronary artery disease, serum zinc level is lower than healthy ones. In a study done by Kazemi et al in 2007, it was observed that serum zinc level were significantly lower in coronary artery patients (16). These findings was also found in the present study, so that serum zincs level in the patients without coronary artery occlusion and without CSFP were significantly higher than other groups. Regarding the high prevalence of these micronutrients' deficiency in the community and considering the pathophysiology of the slow flow of coronary arteries, it seems necessary to conduct further studies in this field.

This study was confronted with various limitations, including a small sample size.

Moreover, in the current study, coronary obstructive disease and the slow flow of the epicardial microvessels were measured visually and dependent on the physician. However, it was tried to carry out all angiographies by only one cardiologist to avoid errors and bias in the results. Moreover, the serum level of zinc and magnesium were in normal range for all patients. Performing studies on more patients with zinc and magnesium deficiency and comparing their findings with people with normal serum level of these trace elements could be a recommendation for future works.

Conclusion

In the present study, no significant relationship was found between the serum level of zinc and magnesium with the intensity of coronary artery slow flow based on TIMI. However, the serum level of zinc was significantly higher in CAD and slow flow negative patients. Further studies seem to be needed to other aspects of this relationship.

Conflicts 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.

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