The Potential of Magnesium Sulfate to Change Serum Lysyl Oxidase and Nitrite Levels in Patients with Atherosclerosis: A Double-Blind Clinical Trial Study

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Introduction

Endothelial dysfunction plays an important role in atherosclerosis plaque formation and coronary artery disease (CAD) (1,2). During CAD, endothelial dysfunction and high LDL-C (low-density lipoprotein cholesterol) concentrations reduce nitric oxide (NO) bioavailability and the expression of an important enzyme called lysyl oxidases (LOX) in endothelial cells (3,4).

Studies suggest that LOX downregulation causes endothelial dysfunction and instability and rupture of atherosclerotic plaques (4-6). In addition, reduction of NO bioavailability increases oxidative stress leading to endothelial dysfunction (1,7).

It has been demonstrated that serum Mg levels are inversely associated with the risk of CAD (8). The results have shown that 53% of cardiac patients are hypomagnesaemic (9,10). In fact, Mg deficiency can promote oxidative and nitrosative stress and, consequently, endothelial dysfunction in CAD patients. Based on previous reports, Mg2+ treatment can potentially protect people against CAD by enhancing endothelium-dependent vasodilation and reducing vascular resistance.
and oxidative stress (8).

Although different mechanisms have been proposed for the cardioprotective role of Mg, its exact mechanism of effect is unclear. Therefore, further investigations are required to explore other possible functions of this important element. In previous works, the effect of Mg on LOX, as a key enzyme involved in endothelial dysfunction, has not been studied. Thus, our research aimed to investigate the treatment effects of MgSO4 on serum LOX levels in patients with moderate CAD. We also studied the effect of Mg administration on serum nitrite and homocysteine levels, as possible mechanisms through which magnesium may control LOX levels.

Materials and Methods

Trial design

This double-blind randomized placebo-controlled clinical trial has a parallel design with a 1:1 allocation ratio (38 people in the placebo group and 38 participants in the Mg-treated group). This investigation was carried out to evaluate the effect of MgSO4 on serum LOX, homocysteine, and nitrite levels in patients with moderate CAD. This randomized study has been approved by the Iranian Registry of Clinical Trials with the code IRCT20151028024756N3. The trial diagram is depicted in Figure 1.

Participants

In this trial, 76 male and female patients aged more than 55 and 50 years, respectively, with moderate CAD and at least one major coronary artery with less than 60% stenosis were included. All subjects underwent angiography via the femoral artery. Subjects with hypertension and hyperlipidemia and smokers were also included.

Pregnant women, patients who were taking magnesium and calcium dietary supplements, those with renal failure (creatinine levels ≥ 1.3 mg/dL in women and ≥ 1.5 in men), increased hepatic enzymes, recent infections, chronic inflammatory diseases, cerebrovascular accidents (CVAs), cancer, and drug sensitivity and patients with hyperthyroidism and hypothyroidism were excluded from this study. All subjects were selected from the angiography center of Shahed Mohammad Marjani hospital, Hormozgan University of Medical Sciences, Bandar Abbas.

Study protocol, Interventions and sample size calculation

The participants were randomly asked to use either capsules containing MgSO4 (300 mg) or placebo capsules daily for 6 months along with their routine treatments, including metoprolol, aspirin, clopidogrel, nitrate, atorvastatin, losartan, and metformin. The MgSO4 supplement and placebo were made by Niak Pharmaceuticals Company (Gorgan, Iran). Both capsules had the same color and were oval in shape, with the placebo capsule containing wheat bran instead of MgSO4. Evaluation of pericardial arteries was done by angiography. According to the number of atherosclerotic vessels, the patients were assigned to four groups, Mg-treated participants with one atherosclerotic vessel (Mg-VR1, n = 14), Mg-treated participants with two atherosclerotic vessels (Mg-VR2, n = 14), placebo participants with one atherosclerotic vessel (Placebo-VR1, n = 14), and placebo participants with two atherosclerotic vessels (Placebo-VR2, n = 14).

Materials and Methods

Figure 1. Study follow diagram
two atherosclerotic vessels (Mg-VR2, n = 17), and two
groups of placebo-treated participants, with one and
two atherosclerotic vessels (placebo-VR1, n = 14 and
placebo-VR2, n = 17) (Figure 1). In this study, computer-
generated random numbers were used to generate the
random allocation sequence, and block randomization
was used with block size = 2 (the two groups consisted of
patients with one atherosclerotic vessel and patients with
two atherosclerotic vessels). Neither the participant nor
the investigator was aware of the capsule contents, but
one of our colleagues had access to the data and patient
information. In this study, the formula for determining
the sample size was \( n = \frac{Zp}{2} \left( \frac{Zp}{2} + Z\beta \right) \frac{d^2}{2} \) (\( Zp = 1.96, Z\beta = 0.84, d = 0.05, SD = 0.012 \)).

**Exclusion and inclusion criteria**

Women and men with moderate CAD aged more than
55 years and 45 years, respectively, were included in
the study, even if they had hypertension or hyperlipidemia or
were smokers.

Pregnant women, patients who were taking magnesium
and calcium dietary supplements, those with renal failure
(serum creatinine levels ≥ 1.3 mg/dL in women and ≥ 1.5
mg/dL in men), increased hepatic enzymes, recent
infections, chronic inflammatory diseases, CVAs, cancer,
and drug sensitivity and patients with hyperthyroidism
or hypothyroidism were excluded from this study.

**Study outcomes**

The primary outcomes, including angiography, lipid
profile, and homocysteine, were measured at the baseline
and 3 and 6 months following magnesium therapy.
The secondary outcome of the study was the levels of
nitrite, LOX, serum glutamate pyruvate transaminase
(SGPT), serum glutamate oxaloacetate transaminase
(SGOT), creatinine (Cr), and blood urea nitrogen (BUN),
measured at the baseline and at the 3 and 6 months
following magnesium therapy.

**Biochemical measurements**

Fasting blood samples were collected three times, before
intervention and at the 3- and 6-month follow-ups. Samples
were centrifuged at 1500 g for 10 minutes. Serum
was separated and maintained at -80 °C for biochemical
assessment. Serum total cholesterol, triglycerides
(TG), Cr, BUN, and homocysteine were measured by
enzymatic methods (Pars Azmun kit, Iran). Very low-
density lipoprotein cholesterol (VLDL-C) concentration
was calculated. Serum LOX was determined by an
ELISA assay (ZellBio GmbH, Germany, Cat. No: ZB-
15122C-H9648). Serum nitrite levels were assessed using
a standard kit (Promega Corporation, USA, G2930 ).
Also, liver function tests, including SGPT and SGOT,
were carried out using available commercial kits (Pars
Azmun kit, Iran).

**Statistical analysis**

The findings were reported as mean±SD, and
Kolmogorov-Smirnov test was used to check the
normality of all variables. Comparisons between groups
and the possible differences between the means of each
group during the intervention were analyzed using one-
way ANOVA and Tukey’s post-hoc test. SPSS software
version 13 was used for the analyses, and P<0.05 was
taken as statistically significant.

**Results**

In the present study, 76 patients were selected according
to the number of atherosclerotic vessels and were randomly
divided into Mg and placebo groups: M-treated group
with one atherosclerotic vessel (Mg-VR1, n = 14), MgSO4-
treated group with two atherosclerotic vessels (Mg-VR2,
n = 17) and two placebo-treated groups with one or two
atherosclerotic vessels (placebo-VR1, n = 14 and placebo-
VR2, n = 17, respectively (Figure 1).

**Effects of MgSO4 on lipid profile, BUN, Cr, and liver enzymes**

**Measurements at baseline**

The baseline total cholesterol, TG, and VLDL-C
concentration are shown in Figure 2. Total cholesterol,
TG, and VLDL-C levels were higher in participants
with two atherosclerotic vessels compared with one
atherosclerotic plaque at the beginning of the study
(\( P<0.001 \)) (Figure 2). We did not observe any change in
the serum levels of BUN, Cr, SGOT, and SGPT at baseline
(Table 1).

**Measurements after Mg treatment**

The effects of 3 and 6 months Mg administration
on serum analyses in different groups are shown in
Figure 2 and Table 2 and 3. Total cholesterol was
significantly decreased 6 months after intervention in
both placebo and Mg-treated groups (\( P<0.001 \)). In
addition, TG decreased at the end of 6 months in the
placebo groups (\( P<0.01 \)), but the plasma TG levels of
Mg-VR2 patients increased after 3 months and then
decreased at 6 months follow up (\( P<0.001 \)).

In patients with one atherosclerotic vessel, MgSO4 was
able to reduce VLDL-C levels 3 months after intervention
(\( P<0.001 \)), but in placebo groups, this reduction took
place after 6 months (\( P<0.01 \)). Therefore, Mg treatment
decreased VLDL-C faster compared with the placebo-
VR1 group (Figure 2).

Following Mg administration, we observed no change
in the serum levels of BUN, Cr, SGOT, and SGPT (Tables 2 and 3).

**Effects of MgSO4 on homocysteine levels**

**Measurement at baseline**

The baseline homocysteine concentrations are shown in
Table 1. Demographic information in MgSO₄ (Mg) treated moderate coronary artery disease patients with one (Mg-VR1) and two (Mg-VR2) atherosclerotic vessels as well as in placebo-treated patients with one (placebo-VR1) and two (placebo-VR2) atherosclerotic vessels before intervention

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mg-treated</th>
<th>Placebo</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (y)</td>
<td>60.07 ± 3.2</td>
<td>60.4 ± 2.1</td>
<td>57.06 ± 2</td>
</tr>
<tr>
<td>Gender (F/M)</td>
<td>7/7</td>
<td>8/9</td>
<td>6/8</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>25.9 ± 1.6</td>
<td>23.8 ± 0.7</td>
<td>25.1 ± 1.1</td>
</tr>
<tr>
<td>SBP (mm Hg)</td>
<td>121.4 ± 4.1</td>
<td>127.8 ± 3.5</td>
<td>130.3 ± 3.5</td>
</tr>
<tr>
<td>DBP (mm Hg)</td>
<td>72.1 ± 1.3</td>
<td>75.8 ± 2.3</td>
<td>78.6 ± 2.1</td>
</tr>
<tr>
<td>HR</td>
<td>71.3 ± 1.57</td>
<td>69.2 ± 1.7</td>
<td>68.8 ± 2.5</td>
</tr>
<tr>
<td>SGOT (U/L)</td>
<td>19.9 ± 1.8</td>
<td>23.1 ± 2.7</td>
<td>20.4 ± 1.1</td>
</tr>
<tr>
<td>SGPT (U/L)</td>
<td>25.6 ± 4.3</td>
<td>28.1 ± 3.6</td>
<td>23.1 ± 3.7</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>14.5 ± 0.7</td>
<td>15.5 ± 0.5</td>
<td>14.6 ± 0.5</td>
</tr>
<tr>
<td>Cr (mg/dL)</td>
<td>0.8 ± 0.01</td>
<td>0.9 ± 0.02</td>
<td>0.8 ± 0.03</td>
</tr>
</tbody>
</table>

BMI (body mass index), SBP (systolic blood pressure), DBP (diastolic blood pressure), HR (heart rate), Cr (creatinine), BUN (blood urea nitrogen), SGPT (serum glutamate-pyruvate transaminase), SGOT (glutamate oxaloacetate transaminase).

Data were presented as mean ± SD and analyzed using one-way ANOVA.

Table 2. Study variables in MgSO₄ (Mg) treated moderate coronary artery disease patients with one (Mg-VR1) and two (Mg-VR2) atherosclerotic vessel as well as in placebo treated patients with one (placebo-VR1) and two (placebo-VR2) atherosclerotic vessels after 3 months of intervention

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mg-treated</th>
<th>Placebo</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>SGOT (U/L)</td>
<td>20.3 ± 1.2</td>
<td>21.1 ± 1.5</td>
<td>21.1 ± 1.2</td>
</tr>
<tr>
<td>SGPT (U/L)</td>
<td>21.6 ± 1.8</td>
<td>22.5 ± 2.4</td>
<td>22.7 ± 1.7</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>12.2 ± 0.7</td>
<td>13.2 ± 0.6</td>
<td>13.6 ± 1.1</td>
</tr>
<tr>
<td>Cr (mg/dL)</td>
<td>0.8 ± 0.02</td>
<td>0.8 ± 0.04</td>
<td>0.7 ± 0.03</td>
</tr>
</tbody>
</table>

Cr (creatinine), BUN (blood urea nitrogen), SGPT (serum glutamate-pyruvate transaminase), SGOT (glutamate oxaloacetate transaminase).

Data was presented as mean ± SD and analyzed using one-way ANOVA.

Figure 2. Serum cholesterol (a), triglyceride (TG, b) and very low density lipoprotein cholesterol (VLDL-C, c) levels in four groups of moderate coronary artery disease (CAD): 1) MgSO₄-treated CAD participants with one atherosclerotic vessel (Mg-VR1), 2) Mg- treated CAD patients with two atherosclerotic vessels (Mg-VR2), 3) placebo-treated CAD patients with one atherosclerotic vessel (placebo-VR1), and 4) placebo-treated CAD patients with two atherosclerotic vessels (placebo-VR2). n = 14 for one atherosclerotic vessel in each group and n = 17 for two atherosclerotic vessels in each group. Data were presented as mean ± SD and analyzed using one-way ANOVA and Tukey’s post-hoc test. * P < 0.001 significant differences compared with the VR1 groups before intervention (base). & P < 0.001 significant difference between the same group 3 months after intervention (3-mt). @ P < 0.001 significant difference compared with the placebo-VR1 group after 3 months (3-mt). # P < 0.001 significant difference compared with the VR1 group 6 months after intervention (6-mt). & P < 0.01 significant difference compared with the placebo-VR1 group 6 months after intervention (6-mt).

Figure 3a. The serum levels of homocysteine were higher in VR2 patients compared to VR1 patients in both the Mg and placebo groups (P < 0.001) (Figure 3a).

Measurement after Mg treatment
Homocysteine variations after 3 and 6 months MgSO₄ treatment and its comparison with placebo groups are shown in Figure 3a.

Decrease of serum homocysteine levels was seen in all groups during the study but the reduction was greater in the placebo groups compared to the Mg- treated groups after 3 months (P < 0.001) (Figure 3a).
Influence of magnesium sulfate on serum lysyl oxidase in atherosclerosis

Table 3. Study variables in MgSO₄ (Mg)-treated moderate coronary artery disease patients with one (Mg-VR1) and two (Mg-VR2) atherosclerotic vessels and placebo-treated patients with one (placebo-VR1) and two (placebo-VR2) atherosclerotic vessels after 6 months of intervention

<table>
<thead>
<tr>
<th>Variables</th>
<th>Mg-treated VR1 (n = 14)</th>
<th>VR2 (n = 17)</th>
<th>Placebo VR1 (n = 14)</th>
<th>VR2 (n = 17)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>SGOT (U/L)</td>
<td>20.1 ± 1.4</td>
<td>19.3 ± 1.1</td>
<td>24 ± 2.6</td>
<td>21.1 ± 0.7</td>
<td>0.2</td>
</tr>
<tr>
<td>SGPT (U/L)</td>
<td>22.5 ± 3.1</td>
<td>22.7 ± 2.1</td>
<td>29.5 ± 6.7</td>
<td>23.2 ± 2.1</td>
<td>0.2</td>
</tr>
<tr>
<td>BUN (mg/dL)</td>
<td>12.1 ± 0.8</td>
<td>12.1 ± 0.8</td>
<td>13.6 ± 0.9</td>
<td>11.8 ± 0.7</td>
<td>0.4</td>
</tr>
<tr>
<td>Ca (mg/dL)</td>
<td>0.8 ± 0.04</td>
<td>0.9 ± 0.03</td>
<td>0.8 ± 0.03</td>
<td>0.9 ± 0.03</td>
<td>0.46</td>
</tr>
<tr>
<td>Cr (creatinine)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>BUN (blood urea nitrogen)</td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>SGPT (glutamate-pyruvate transaminase)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>SGOT (serum glutamate-pyruvate transaminase)</td>
<td></td>
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</tr>
</tbody>
</table>

Cr (creatinine), BUN (blood urea nitrogen), SGPT (serum glutamate-pyruvate transaminase), SGOT (glutamate oxaloacetate transaminase).

Data were presented as mean ± SD and analyzed using one-way ANOVA.

Figure 3. Serum homocysteine (a), lysyl oxidase (b) and nitrite (c) levels in MgSO₄ (Mg)-treated moderate coronary artery disease patients with one atherosclerotic vessel (Mg-VR1), Mg-treated moderate coronary artery disease patients with two atherosclerotic vessels (Mg-VR2), placebo-treated moderate coronary artery disease patients with one atherosclerotic vessel (placebo-VR1) and placebo-treated moderate coronary artery disease patients with two atherosclerotic vessels (placebo-VR2). n = 14 for one atherosclerotic vessel in each group and n = 17 for two atherosclerotic vessels in each group. Data were presented as mean ± SD and analyzed using one-way ANOVA and Tukey’s post-hoc test. * P < 0.001 significant difference compared with VR1 groups before intervention (baseline). $ P < 0.001 significant difference compared with placebo-VR1 group 3 months after intervention (3-mt). @ P < 0.001 significant difference with the placebo-VR1 group 6 months after intervention (6-mt). # P < 0.001 significant difference compared with the placebo-VR2 group 3 months after intervention (3-mt).

Effects of MgSO₄ on serum LOX levels
Measurement at baseline
The baseline LOX concentrations are shown in Figure 3b. There was no significant difference in LOX levels among the four groups (Figure 3b).

Measurement after Mg treatment
LOX changes after 3 and 6 months MgSO4 treatment are shown in Figure 3b.

Three months after treatment, serum LOX was found to be higher in the VR1 and VR2 Mg-treated groups than in the placebo groups, so Mg was able to increase LOX levels (P < 0.001). Moreover, the LOX levels returned to baseline after 6 months in the Mg-treated groups (Figure 3b).

Effects of MgSO₄ on serum levels of nitrite
Measurement at baseline
The baseline serum nitrite concentrations are shown in Figure 3c. Nitrite levels were lower in the VR2 patients compared with the VR1 group in both Mg- and placebo-treated groups (P < 0.001) (Figure 3c).

Measurement after Mg treatment
Alteration in serum levels of nitrite after 3 and 6 months treatment with MgSO₄ are shown in Figure 3c.

A rise of nitrite levels was seen in the VR2 groups in both intervention and placebo groups after 3 months, and it then returned to the baseline levels after 6 months (P < 0.001). This enhancement of nitrite levels in the placebo-VR2 group was significantly greater than the Mg-VR2 group after 3 months (P < 0.001), indicating Mg had prevented the increase in nitrite levels in VR2 patients (Figure 3c).

Discussion
In this study, we found that Mg was able to maintain LOX levels and reduce VLDL-C, homocysteine and nitrite, especially in the 3 months after intervention. The LOX enzyme may be considered an effective modifier of Mg’s influence on cardiovascular disorders. These observations demonstrated that Mg can prevent the decrease of LOX levels and explain the beneficial effects of this element in CAD.

Maintaining the elasticity of the vascular system via regulation of collagen and elastin turnover is one way Mg protects vessels against vessel stiffness (11). On the other hand, the development of vessel walls is induced by LOX (12). Thus, it is reasonable to assume that Mg and this enzyme have a common pathway to regulate vessel wall development and elasticity. The impairment of the NO system usually accompanies cardiovascular diseases...
(CVDs). The reduction of NO production is associated with endothelial dysfunction and atherosclerosis (13,14). Our data showed that Mg was able to maintain LOX levels and decrease nitrite levels. Nitrite assay is considered an index of NO synthesis (15). Previous studies have demonstrated that elastic fiber formation occurs via NO production. Therefore, decrease in NO synthesis could be responsible for the reduction of vessel elasticity and induction of plaque formation (16). It is suggested that NO accelerates the expression of LOX, which induces filament cross-links (17). In addition, Mg stimulates NO production and endothelial NOS (eNOS) activity, so it could be postulated that in this way, Mg can induce cardiovascular protection (11). One study indicated that Mg alters LOX function, but the underlying mechanism needs further investigation (11). As a vasodilator, Mg contributes to vessel function (18). In physiological conditions, NO production by eNOS accounts for the biological function of vessel strength. However, NO production by inducible NOS (iNOS) accelerates oxidative stress, leading to initiation and progression of cardiovascular problems (19,20). In addition, increase the activity of iNOS, and eNOS impairment appears to play a fundamental role in the progression of atherosclerosis (19,21). We observed that nitrite levels were significantly greater in the placebo-VR2 group than the Mg-VR2 group after 3 months. This means Mg may prevent nitrite levels from increasing by inhibiting iNOS expression in patients with high severity of atherosclerosis, modulating LOX activity and improving CAD.

Epidemiological studies have shown that a rise in plasma homocysteine can induce atherosclerotic plaque formation as an independent risk factor. Hyperhomocysteinemia accelerates thrombogenicity, oxidative stress status, and endothelial dysfunction (22,23). Even slightly increased serum homocysteine levels can be a risk factor for the development of coronary disease (24). Moreover, optimal homocysteine levels are below 10 to 12 μmol/L, and the initial threshold level for hyperhomocysteinemia can be 15 μmol/L (25). In our study, subjects with two atherosclerotic vessels showed significantly higher basal concentrations of homocysteine than subjects with one atherosclerotic vessel, indicating that homocysteine level is associated with the degree of CAD. In line with our research, the result of a large cohort study in patients undergoing coronary angiography has revealed that elevated homocysteine is significantly associated with the degree of CAD (22). In our research, three and 6 months after intervention, the level of homocysteine significantly decreased both in patients receiving placebo and MgSO4, compared to baseline levels. However, the reduction was greater in placebo groups than Mg-treated groups after 3 months. In line with the current study, Mg treatment has been reported to reduce homocysteine level in pregnancy-induced hypertension syndrome (26). It is assumed that Mg administration can modulate homocysteine levels in CAD, because the major enzymes of homocysteine metabolism pathway are Mg-dependent (26). Our study demonstrated that in 3 months of treatment, MgSO4 was able to maintain LOX at high levels, and after 6 months, the LOX levels returned to baseline. It has been shown that disturbance in LOX expression and activity is associated with cardiovascular problems and the normal activity of LOX is necessary for vascular hemostasis (3,27). The data showed that LOX deficiency leads to alternation of endothelial function and vascular wall remodeling (28). Moreover, in the initial phase of CAD, the downregulation of LOX promotes cardiovascular complications (3). Additionally, overactivity of LOX has been reported in hypertension and chronic CAD (29). In the present work, the enhancement of LOX levels in the initial phase of Mg treatment may lead to the amelioration of atherosclerosis. Hyperhomocysteinemia has been shown to interfere with post-translational modifications of collagen, directly by inhibiting LOX and indirectly by downregulation of mRNA expression of LOX and other genes that are involved in collagen cross-linking (30). Based on our data, serum homocysteine levels in the magnesium groups showed a slower decrease compared to the placebo groups, especially after 3 months. In fact, in the group that received magnesium for 3 months, the reduction of homocysteine level kept it in the optimum range. At the same time, LOX was found to be higher in the VR1 and VR2 Mg-treated groups than in the placebo groups, suggesting that Mg was able to maintain LOX levels. Based on these results, we assume Mg can modulate LOX levels by controlling serum homocysteine concentration. Also, participants (both in placebo and Mg groups) received their routine treatments, including atorvastatin, aspirin, clopidogrel, metoprolol, nitrate, losartan, and metformin, so we assume the reduction of homocysteine levels is related to these drugs in the placebo and mg groups.

Hyperlipidemia is an early event that accelerates CAD development and can alter the composition and permeability of the endothelial barrier. Hypercholesterolemia can alter the endothelial extracellular matrix and induce endothelial dysfunction by significant downregulation of vascular LOX expression (3,27). Our results demonstrates that cholesterol and TG levels were significantly decreased 6 months after intervention in both the placebo and Mg-treated groups. Also, in patients with one atherosclerotic vessel, Mg decreased VLDL-C faster compared with placebo-VR1. Also we assume that Mg can appropriately maintain LOX levels in patients with a lower degree of CAD by lowering the VLDL levels.

In order to show the safety of Mg-treatment, liver and kidney function tests were also conducted. Serum Cr, BUN, SGOT, and SGPT activity showed no significant
difference between the Mg-treated and placebo-treated groups, and liver and kidney function were both within normal range. Therefore, based on our results, magnesium administration for 6 months does not show any toxic effect on the kidney or liver.

Conclusion
Our findings showed that Mg administration in CAD patients for 6 months was able to modulate LOX levels and decrease serum levels of some factors that affect the expression and activity of LOX, including VLDL-C, homocysteine, and nitrite. Therefore, Mg may be considered as an auxiliary supplement to improve some risk factors of CAD by modulating LOX levels.

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Authors’ Contribution
Data curation: Fariba Azarkish, Nepton Soltani, Aghdas Dehghani.
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Investigation: Fariba Azarkish, Nepton Soltani, Aghdas Dehghani.
Project administration: Fariba Azarkish, Aghdas Dehghani.
Supervision: Fariba Azarkish, Nepton Soltani, Aghdas Dehghani.
Writing–original draft: Fariba Azarkish, Aghdas Dehghani.
Writing–review & editing: Fariba Azarkish, Nepton Soltani, Ebrahim Eftekhar, Hossein Farshidi, Seyed Alireza Sobhani, Mahdiye Eslami, Aghdas Dehghani.

Competing Interests
The authors declare that they have no competing interests.

Consent for Publication
All of the authors have consented to publication.

Data Availability Statement
The datasets are available from the corresponding author on reasonable request.

Ethical Approval
The study was approved by the Ethics Committee of Hormozgan University of Medical Sciences with the reference number IR.HUMS.REC.1397.308, approval date 2019-02-17. Informed written consent to participate in the study was obtained from all subjects.

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