Low Serum Magnesium Predicts Neurological Events in Patients With Advanced Atherosclerosis

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Background and Purpose—Magnesium (Mg) deficiency is thought to be a risk factor for cerebrovascular atherosclerosis and complications. We investigated the prognostic impact of Mg serum levels with respect to the occurrence of neurological events in patients with advanced atherosclerosis.

Methods—We prospectively studied 323 patients with symptomatic peripheral artery disease and intermittent claudication (197 men; median age, 68 years). Serum Mg was determined, and patients were followed for a median of 20 months (interquartile range, 12 to 25 months) for the occurrence of neurological events, defined as ischemic stroke and/or carotid revascularization (carotid endarterectomy or carotid stenting). Multivariate Cox proportional hazards analysis was applied to assess the association of serum Mg (in tertiles) and neurological events.

Results—Neurological events occurred in 35 patients (11%) (15 patients with stroke, 13 with carotid revascularization, and 7 with stroke and subsequent revascularization). Compared with patients in the highest tertile of Mg serum levels (>0.84 mmol/L), patients with Mg serum values <0.76 mmol/L (lowest tertile) exhibited a 3.29-fold increased adjusted risk (95% CI, 1.34 to 7.90; \( P = 0.009 \)) for neurological events, but patients with Mg serum values of 0.76 mmol/L to 0.84 mmol/L (middle tertile) had no increased risk (adjusted hazard ratio, 1.10; 95% CI, 0.35 to 3.33; \( P = 0.88 \)). Mg serum levels were not associated with all-cause mortality (\( P = 0.87 \)) or coronary events (\( P = 0.67 \)) during follow-up.

Conclusions—Low Mg serum levels indicate an increased risk for neurological events in patients with symptomatic peripheral artery disease, favoring Mg substitution therapy in those patients with advanced atherosclerosis. (Stroke. 2004;35:22-27.)

Key Words: atherosclerosis ■ magnesium ■ risk factors ■ stroke

Magnesium, a natural calcium antagonist,\(^1\)\(^\text{,}^2\) modulates vasomotor tone, blood pressure, and peripheral blood flow.\(^3\) Serum contains a small proportion of total body Mg, partially bound to proteins, in complex with small anion ligands and as free ionized Mg.\(^3\) The small proportion of ionized Mg in the circulation is thought to exert several beneficial effects on vascular endothelium and function.\(^4\)

Mg deficiency was shown to trigger vasoconstriction and enhance vascular endothelial injury, thus promoting the development and progression of atherosclerosis.\(^5\) In this context, numerous studies elucidated a significant depression of Mg serum levels in patients with vascular diseases.\(^6\)\(^\text{-}^\text{10}\) The largest of these trials, the Atherosclerosis Risk in Communities Study (ARIC),\(^6\)\(^\text{,}^7\) demonstrated an inverse relation of serum and dietary Mg and the development of carotid atherosclerosis in healthy middle-aged adults. However, it is indeterminate whether low serum Mg also promotes the occurrence of cerebrovascular adverse events in patients with preexisting, advanced atherosclerosis.

Patients with symptomatic peripheral artery disease (PAD) are at high risk for complications of atherosclerotic disease.\(^1\)\(^\text{,}^1\)\(^\text{,}^1\) In particular, a high incidence of concomitant cerebrovascular lesions predisposes these patients to neurological adverse events. We hypothesized that low Mg serum levels are associated with an increased incidence of neurological adverse events in patients with symptomatic PAD. Therefore, the aim of the study was to investigate the association of Mg and a combined neurological end point consisting of ischemic stroke and carotid endarterectomy or stenting in patients with intermittent claudication.

Subjects and Methods

Study Design

The study was designed as a prospective cohort study. We included all consecutive patients with symptomatic PAD and intermittent claudication who were admitted to the Angiology Department of a tertiary care university hospital from March 1, 2000, to March 1, 2003.
Patient Data
At admission, patients’ medical history and data from physical examination were recorded on a standard questionnaire by 2 independent observers. Clinical history and physical examination were evaluated with special attention to cardiovascular risk factors and comorbidities: age, sex, smoking habits, hyperlipidemia, arterial hypertension, diabetes mellitus, coronary artery disease, history of cerebrovascular events, and current medication. All patients were taking antithrombotic drugs (aspirin and/or clopidogrel). Data were checked for interobserver agreement at the day of patients’ discharge. In case of discrepancies, both investigators in consensus reevaluated the patient.

Laboratory Parameters
A complete series of routine laboratory investigations, including glycosylated hemoglobin (HbA1c), LDL and HDL cholesterol, complete blood cell count, and serum creatinine, was performed. Antecubital venous blood samples for determination of serum Mg values were taken at admission. Serum Mg concentration was determined on a Roche/Hitachi MODULAR analyzer with the use of the xylidyl blue reaction according to the manufacturer’s instructions. The reference value is 0.7 to 1.0 mmol/L, and the intratest coefficient of variation is 1.2%.

Study End Points
The primary study end point was the occurrence of neurological events during follow-up, defined as major or minor ischemic stroke and carotid endarterectomy (CEA) or carotid artery stenting (CAS). Mandibular cranial CT was used for confirmation of the diagnosis and to differentiate between ischemic versus hemorrhagic strokes. Furthermore, death from any cause and other cardiovascular events (myocardial infarction, coronary artery bypass graft, percutaneous coronary interventions) were considered secondary objectives.

Follow-Up
Patients were clinically reevaluated routinely 3, 6, and 12 months after hospital discharge and thereafter annually at the outpatient ward of our department. For further evaluation of neurological events, a follow-up questionnaire was sent to each patient. Information from the follow-up questionnaire was validated by reviewing the hospital discharge reports of readmissions due to neurological events. If the follow-up questionnaire was not returned, personal telephone contact with the patients, their relatives, or the treating physicians was established. Further information was obtained by reviewing the hospital discharge reports of any other readmission during the follow-up period. Presumed causes of stroke were classified according to the Trial of Org 10172 in Acute Stroke Treatment (TOAST) classification13 on the basis of the findings from clinical examination, CT, echocardiography (transthoracic), and carotid ultrasound evaluation.

Definitions
The diagnosis of PAD was assessed by clinical evaluation, oscillography, ankle-brachial index measurements, and duplex sonography and confirmed by lower limb angiography. For categorization of PAD, the Fontaine classification was used to differentiate patients with PAD stage Ia (maximum walking distance >200 m) versus PAD stage Ib (maximum walking distance <200 m). Diabetes mellitus was defined as fasting blood glucose levels >110 mg/dL, measured 3 times, pathological oral glucose tolerance tests, or HbA1c >6.5% and was considered to be present in all patients taking antidiabetic medication. Hyperlipidemia was defined as fasting total serum cholesterol >200 mg/dL, LDL cholesterol >130 mg/dL, or serum triglycerides >180 mg/dL and was considered present in all patients receiving lipid-lowering therapy (statins were used routinely at our institution). Arterial hypertension was diagnosed according to World Health Organization criteria. Patients who were smoking >3 cigarettes per day were regarded as current smokers. Coronary artery disease was classified according to the Canadian Cardiovascular Society classification, and routine evaluation included treadmill exercise testing, dobutamine echocardiography, myocardium scintigraphy, and coronary angiography in selected cases.

Statistical Analysis
Continuous data are presented as median and interquartile range (IQR) (range, 25th to 75th percentile). Discrete data are given as counts and percentages. We used χ2 tests to compare proportions and Mann-Whitney U tests for univariate comparison of continuous data. Event-free survival rates according to patients’ baseline serum Mg level (in tertiles) are presented as a Kaplan-Meier curve and compared by means of the log rank test. Multivariate Cox proportional hazards analysis was applied to assess the effect of Mg on event-free survival. Baseline variables were entered as possible predictor variables into the model to adjust for confounding effects if (1) they were imbalanced between patients with low serum Mg (lowest tertile) compared with patients in the middle and highest tertiles of serum Mg, indicated by a probability value <0.2, or (2) they were imbalanced between patients with and without neurological events, indicated by a probability value <0.2. The decision to identify possible confounders by comparing the lowest tertile of Mg versus the remaining patients was made post hoc on the basis of the finding that these patients had an increased risk for neurological events. We tested for interactions between baseline variables by stratification as well as multiplicative interaction terms and log likelihood χ2 tests. Results of the Cox logistic regression model were presented as hazard ratio (HR) and 95% CI. A 2-sided probability value <0.05 was considered statistically significant. Calculations were performed with SPSS for Windows (version 10.0, SPSS Inc) and Stata (release 8.0).

Results
Patients
We studied 323 of 338 patients (96%) who were admitted with symptomatic PAD and intermittent claudication during the study period. Fifteen patients (4%) had to be excluded because of missing follow-up data. The median age of the 323 patients was 68 years (IQR, 58 to 76 years), and 197 patients were male (61%). The water supply in the area where the study was undertaken is rated “soft,” indicated by a value of 9.80 dH, which is in the lowest tertile of degrees of German hardness (dH) for drinking water (reference range, 5 to 30 dH).

Serum Magnesium
Serum Mg at admission was a median of 0.81 mmol/L (IQR, 0.74 to 0.85 mmol/L). For further analysis Mg values were divided into tertiles: lowest tertile, Mg <0.76 mmol/L; middle tertile, Mg 0.76 to 0.84 mmol/L; highest tertile, Mg >0.84 mmol/L. Cardiovascular risk factors and comorbidities in patients with low Mg (lowest tertile) versus patients in the middle and highest Mg tertiles were equally balanced (Table 1) with the exception of diabetes mellitus and clinical stage of coronary artery disease: patients with diabetes had significantly lower Mg levels compared with patients without diabetes. Furthermore, slightly lower Mg levels were found in patients with a higher clinical stage of coronary artery disease. The use of diuretics was considered the most important single confounder because it may influence hypertension as a risk factor of stroke as well as the Mg serum level. Thus, without adequate adjustment any effect of Mg on stroke risk may be artifactual. Overall, 258 patients (80%) received any diuretic drug, in 195 patients arterial hypertension was...
considered the primary cause of its use, and in 63 patients congestive heart failure was considered the primary cause of its use. As expected, patients receiving diuretics were more frequently in the lowest tertile of serum Mg levels (Table 1).

Follow-Up for Neurological Events
Neurological events occurred in 35 patients (11%): 15 patients suffered a stroke, 5 patients had a stroke and subsequently underwent CAS, 2 patients had an stroke and subsequent CEA, 11 patients underwent CAS, and 2 patients had CEA. All 22 patients with stroke had an ischemic stroke according to the respective hospital discharge reports based on findings of cranial CT. The presumed etiology of stroke according to the TOAST classification was macrovascular in 15 patients, microvascular in 2 patients, cardioembolic in 4 patients, and undetermined in 1 patient. The indications for CAS in 11 patients were rapidly progressive internal carotid artery stenosis from <70% to >90% within 6 months, as indicated by color-coded duplex sonography and confirmed by angiography in 6 patients, and transient ischemic attacks in 5 patients (amaurosis fugax in 4 patients and transient contralateral arm paresthesia in 1 patient). Indications for CEA in 2 patients were transient ischemic attacks in both patients (amaurosis fugax).

Baseline data in patients with and without neurological events are presented in Table 2. As expected, patients with diabetes mellitus as well as patients with a history of stroke or myocardial infarction were more likely to suffer a neurological event. Patients with arterial hypertension and patients with a higher clinical stage of coronary artery disease also had a trend toward a higher frequency of neurological events. Higher serum creatinine was associated with a trend toward increased neurological event rates. Patients receiving statin therapy had a higher incidence of neurological events, which is certainly due to patient selection.

Magnesium and Risk of Neurological Events
Patients with a Mg level <0.76 mmol/L had a significantly increased risk for neurological events during follow-up (P=0.0059), whereas patients in the middle and highest tertiles had a closely comparable event-free survival (Figure). Being aware of several possible confounders, we applied a multivariate Cox proportional hazards model to assess the association of Mg and the primary study end point, adjusting for diabetes mellitus, smoking, serum creatinine (in tertiles), history of myocardial infarction, history of stroke, and use of diuretic and statin therapy. Arterial hypertension was not included simultaneously with "diuretics" because of significant collinearity. Patients with Mg values <0.76 mmol/L exhibited a 3.29-fold increased adjusted risk (95% CI, 1.34 to 7.90; P=0.009) for neurological events compared with patients with higher Mg serum levels (Table 3). Alternatively, when we included arterial hypertension instead of use of diuretics into the multivariate model, a comparable effect size was found (adjusted HR, 3.33; 95% CI, 1.40 to 7.99; P=0.006).

| TABLE 1. Demographic Data, Cardiovascular Risk Factors, and Comorbidities in 327 Patients With Intermittent Claudication According to the Baseline Serum Level of Magnesium (Lower Tertile Versus Middle and Upper Tertiles) |
|---------------------------------|---------------------------------|---------------------------------|---------------------------------|
| | Mg <0.76 mmol/L (n=113, 35%) | Mg >=0.76 mmol/L (n=210, 65%) | P Value |
| Age, y | 69 (58 to 76) | 68 (58 to 76) | 0.87 |
| Male sex | 67 (59%) | 130 (62%) | 0.65 |
| Arterial hypertension | 85 (75%) | 156 (74%) | 0.85 |
| Diabetes mellitus | 59 (52%) | 62 (30%) | <0.001 |
| Smoking | 56 (50%) | 88 (42%) | 0.19 |
| Hyperlipidemia | 92 (81%) | 171 (81%) | 0.99 |
| Serum creatinine, mg/dL | 1.03 (0.90 to 1.27) | 1.07 (0.93 to 1.26) | 0.28 |
| PAD stage Fontaine Iib* | 92 (81%) | 166 (79%) | 0.61 |
| Coronary artery disease (CCS) | | | 0.083 |
| I | 22 (20%) | 59 (28%) | |
| II | 25 (22%) | 33 (16%) | |
| III | 3 (3%) | 1 (1%) | |
| IV | ... | ... | |
| History of myocardial infarction | 25 (22%) | 51 (24%) | 0.66 |
| History of stroke | 10 (9%) | 25 (12%) | 0.40 |
| Serum ionized calcium, mmol/L | 2.39 (2.34 to 2.46) | 2.41 (2.33 to 2.46) | 0.79 |
| Use of diuretic drugs | 102 (90%) | 156 (74%) | 0.001 |
| Statin therapy | 64 (57%) | 116 (55%) | 0.81 |

Data are given as counts and percentages or median and interquartile range (range from the 25th to the 75th percentile)

*Peripheral artery disease with severe claudication (walking distance <200 m).
CCS indicates Canadian Cardiovascular Society.
Magnesium and Risk of Death and Other Cardiovascular Events

During the follow-up period, 29 patients (9%) died. Three of the 29 patients died after a major stroke, and none of the patients died during or after surgical or endovascular treatment of carotid stenosis. Mg serum levels (in tertiles) were not significantly associated with death in this patient sample (log rank $P=0.87$). Coronary events were observed in 34 patients (11%) during the follow-up period: 11 myocardial infarctions, 32 percutaneous coronary interventions, and 5 coronary artery bypass grafts. Mg serum levels (in tertiles) also were not significantly associated with coronary events in these patients (log rank $P=0.67$).

Analysis of Missing Data

The 15 patients with missing follow-up data did not significantly differ with respect to demographic data, clinical characteristics, and baseline Mg levels compared with patients with complete follow-up data. Furthermore, including these patients in either the “event-free survival” or “neurological events” (assuming a 12-month follow-up) categories in the final model did not substantially influence the observed effect sizes. Adjusted HRs for neurological events for the lowest and middle tertiles compared with the highest tertile were 2.91 and 1.09 for the “event-free survival” approach for all 15 patients and 3.19 and 1.25 for the “neurological events” approach for all 15 patients.

Discussion

We found that low Mg serum levels were associated with an increased risk of neurological events in patients with symptomatic PAD. This association in patients with advanced atherosclerosis has not yet been described. Our findings confirm prior observations suggesting a relevant role of Mg in the development and progression of cerebrovascular atherosclerotic disease and complications. However, Mg serum levels were not associated with all-cause mortality or coronary events in this patient sample.

| TABLE 2. Demographic Data, Cardiovascular Risk Factors, and Comorbidities in 327 Patients With and Without Neurological Events During a Median Follow-Up Time of 20 Months (IQR 12 to 25). |
|-----------------|-----------------|--------|
| Event-Free Survival (n=288, 89%) | Neurological Events (n=35, 11%) | $P$ Value |
| Age, y 68 (58 to 76) | 66 (58 to 75) | 0.96 |
| Male sex 175 (61%) | 22 (63%) | 0.81 |
| Arterial hypertension 211 (73%) | 30 (86%) | 0.11 |
| Diabetes mellitus 103 (36%) | 18 (51%) | 0.071 |
| Smoking 127 (44%) | 17 (49%) | 0.62 |
| Hyperlipidemia 232 (81%) | 31 (89%) | 0.25 |
| Serum creatinine, mg/dL 1.05 (0.92 to 1.25) | 1.16 (1.00 to 1.36) | 0.096 |
| PAD stage Fontaine IIb* 230 (80%) | 28 (80%) | 0.99 |
| Coronary artery disease (CCS) | | |
| I 75 (26%) | 6 (17%) | |
| II 47 (16%) | 11 (31%) | |
| III 4 (1%) | ...(-%) | |
| IV ... | ... | |
| History of myocardial infarction 61 (21%) | 15 (43%) | 0.004 |
| History of stroke 27 (9%) | 8 (23%) | 0.016 |
| Serum ionized calcium, mmol/L 2.41 (2.33 to 2.46) | 2.39 (2.20 to 2.45) | 0.37 |
| Use of diuretic drugs 232 (81%) | 26 (74%) | 0.38 |
| Statin therapy 154 (54%) | 26 (74%) | 0.019 |

Data are given as counts and percentages or median and interquartile range (range from the 25th to the 75th percentile).

*Peripheral artery disease with severe claudication (walking distance <200 m).

CCS indicates Canadian Cardiovascular Society.
Mg is one of the most abundant ions in human cells, and its serum concentration is remarkably constant in healthy subjects. However, even small alterations in the extracellular Mg concentration can influence arterial smooth muscle tone in cerebral arteries. Although the measurement of serum Mg does not always reflect the overall status of Mg metabolism, serum Mg correlates well with intracellular free Mg, the physiologically active form of the element. Reduced intracellular levels of Mg have been described in patients with acute myocardial infarction and also are significant as a coregulator of platelet-dependent thrombosis. Therefore, serum Mg is the most practicable and commonly used parameter for assessing disorders of Mg metabolism in clinical routine.

Recently, it has been demonstrated that low plasma levels of Mg accelerate atherogenesis by promoting inflammation and increasing LDL concentration and oxidative modification. In rodents, dietary Mg restriction induces exacerbation of atherogenesis by upregulation of serum cholesterol and triglycerides, stimulation of lipid peroxidation, and increased intimal lipid deposition in the vascular wall. Furthermore, enhanced inflammatory cell recruitment and a release of growth factors that induce cell migration and proliferation were observed after a low-Mg diet. Increased Mg intake was suggested to counteract these effects in animal models.

Epidemiological studies have indicated a direct relation between atherosclerosis and low serum Mg, which, in turn, depends on dietary intake. A direct relation between serum Mg levels and acute neurological events has not yet been described. However, Mg has been demonstrated to coregulate cerebrovascular smooth muscle tone, and low serum Mg was associated with the development of cerebrovascular atherosclerosis. There is some evidence supporting the hypothesis that dietary Mg intake potentially contributes to blood pressure and stroke reduction. In particular, hypertensive individuals were suggested to benefit from increased Mg intake. Furthermore, Mg intake from drinking water was suggested to exert a significant protective effect on the risk of cerebrovascular disease and death from stroke. When these and our findings are considered together, it seems plausible that low Mg serum levels promote neurological events in patients with advanced atherosclerosis. Therefore, the substitution of Mg may be favored in neurologically high-risk patients. However, controversial clinical data exist on the potential benefits of oral Mg substitution. Although Mg substitution was found to improve endothelial function and exercise tolerance in patients with stable coronary artery disease as well as after acute myocardial infarction, others found no significant clinical improvement in patients with Mg therapy. Our data support the view that it at least may be worth considering Mg substitution for patients with low Mg serum levels who are at high risk for neurological events.

Some limitations of the present study must be acknowledged. In particular, the combined study end point of any ischemic stroke and carotid revascularization may include different pathophysiological entities. However, according to the TOAST classification of stroke, the majority of events were due to macrovascular disease. Furthermore, the categories CAS and CEA also include patients with macrovascular disease, suggesting that low Mg is mainly a risk factor for stroke based on macrovascular disease. Nevertheless, considerably larger patient numbers will be necessary to confirm our findings and to distinguish between the different entities of neurological events. Another limitation is the lack of data on other drugs that may influence serum Mg levels, such as biphosphonates or nonsteroidal anti-inflammatory drugs, and dietary Mg intake of the study population.

**Conclusion**

Low serum Mg levels (<0.76 mmol/L) are a risk factor for neurological events in patients with symptomatic PAD, fa-
voring Mg substitution therapy in those patients with advanced atherosclerosis.

References
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