Effect of oral magnesium supplementation on physical performance in healthy elderly women involved in a weekly exercise program: a randomized controlled trial

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ABSTRACT
Background: Magnesium deficiency is associated with poor physical performance, but no trials are available on how magnesium supplementation affects elderly people’s physical performance.
Objectives: The aim of our study was to investigate whether 12 wk of oral magnesium supplementation can improve physical performance in healthy elderly women.
Design: In a parallel-group, randomized controlled trial, 139 healthy women (mean ± SD age: 71.5 ± 5.2 y) attending a mild fitness program were randomly allocated to a treatment group (300 mg Mg/d; n = 62) or a control group (no placebo or intervention; n = 77) by using a computer-generated randomization sequence, and researchers were blinded to their grouping. After assessment at baseline and again after 12 wk, the primary outcome was a change in the Short Physical Performance Battery (SPPB); secondary outcomes were changes in peak torque isometric and isokinetic strength of the lower limbs and handgrip strength.
Results: A total of 124 participants allocated to the treatment (n = 53) or control (n = 71) group were considered in the final analysis. At baseline, the SPPB scores did not differ between the 2 groups. After 12 wk, the treated group had a significantly better total SPPB score (Δ = 0.41 ± 0.24 points; P = 0.03), chair stand times (Δ = −1.31 ± 0.33 s; P < 0.0001), and 4-m walking speeds (Δ = 0.14 ± 0.03 m/s; P = 0.006) than did the control group. These findings were more evident in participants with a magnesium dietary intake lower than the Recommended Dietary Allowance. No significant differences emerged for the secondary outcomes investigated, and no serious adverse effects were reported.
Conclusions: Daily magnesium oxide supplementation for 12 wk seems to improve physical performance in healthy elderly women. These findings suggest a role for magnesium supplementation in preventing or delaying the age-related decline in physical performance. This trial was registered at clinicaltrials.gov as NCT01971424.


INTRODUCTION
Magnesium has a fundamental role in muscle function and is essential to energy metabolism, transmembrane transport, and muscle relaxation and contraction (1–4). Magnesium deficiency has proved capable of impairing exercise capacity and reducing physical performance (5). A large cross-sectional study on older people showed a strong association between serum magnesium and several muscle performance tests (6). Magnesium depletion is also associated with an increased inflammatory state, muscle cell alterations attributable to increased oxidative stress, and impaired intracellular calcium homeostasis (7). All these factors negatively affect muscle mass and function and could exacerbate the sarcopenia typical of old age.

Elderly people are particularly susceptible to magnesium deficiency for several reasons, including an inadequate dietary intake, a less efficient magnesium absorption, and greater losses in stools and urine (2). Elderly women may be more susceptible to magnesium deficiencies than men, partly because they are more likely to have osteoporosis, which limits the exchange of magnesium between bone and blood (8). Older women also have a low magnesium intake in their diet: in Europe, 77% of women older than 65 y have a dietary intake below the Recommended Dietary Allowance (RDA), and 23% ingest less than two-thirds of the RDA (2, 9).

It has been widely accepted that magnesium has a positive effect on muscle function, but studies on the efficacy of magnesium supplementation in young people have generated contrasting results (10–15). Differences in magnesium status may be the reason for these different findings, because some researchers have suggested that magnesium supplementation might only help magnesium-deficient people (16), and many studies did not assess magnesium status (or they did so using scarcely sensitive methods) before providing supplementation. Finally, to the best of our knowledge, no such investigations appear to have been conducted in older people.

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2 No funding was used in the design, implementation, analysis, or interpretation of the data. Magnesium samples were provided free of charge by Sanofi-Aventis.
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4 Abbreviations used: PASE, Physical Activity Scale for the Elderly; PT, peak torque; RDA, Recommended Dietary Allowance; SPPB, Short Physical Performance Battery; 25(OH)D, 25-hydroxyvitamin D.

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Our hypothesis is that oral magnesium supplementation could improve physical performance and muscle function in old women, and the aim of our study was thus to test whether 12 wk of daily oral magnesium supplementation (300 mg Mg) could improve muscle strength and physical performance in a group of healthy elderly women.

SUBJECTS AND METHODS

Participants

This study was conducted at the Geriatrics Department of Padova University between January 2012 and March 2013. Women older than 65 y were recruited on a voluntary basis from among elderly people attending a twice-weekly mild fitness program at public gyms in Padova, Italy.

Candidates were excluded if they had evidence of renal failure, chronic or acute infection, a history or evidence of malignancy in the past 5 y (except for nonmelanoma skin neoplasia), significant cardiovascular or pulmonary diseases, uncontrolled metabolic disease (diabetes, anemia, or thyroid disease), or electrolyte abnormalities or if they used any drugs or supplements that might interfere with magnesium metabolism (magnesium, potassium, calcium, digitalis, or proton pump inhibitors). During a screening visit, their healthy condition was established by trained medical personnel on the strength of their medical history, a clinical examination, and routine biochemical tests (eg, measurement of glucose and electrolyte concentrations, renal and liver function tests, and a complete blood count). Given the close relation between 25-hydroxyvitamin D [25(OH)D] and physical performance in elderly people (17), individuals with 25(OH)D concentrations $\geq 25$ nmol/L ($n = 14$) at the screening visit were supplemented with 800 IU cholecalciferol/d orally, and their concentrations were remeasured after 1 mo. Those whose 25(OH)D concentrations remained $<50$ nmol/L after supplementation ($n = 2$) were ruled out before the randomization.

During the follow-up, all participants continued their fitness program and their attendance was monitored and recorded biweekly in their records. The use of new supplements or medications during the study period was not permitted and was considered a reason for exclusion, but none of the women in the sample met this exclusion criterion.

The study was designed in accordance with the Helsinki Declaration and was approved by the local Ethical Committee (no. 491/2011). All participants were fully informed about the nature, purpose, procedures, and risks of the study and gave their informed consent. Our trial complied with the Consolidated Standards of Reporting Trials statement for randomized trials (Figure 1) (18).

Randomization, intervention, and allocation

Volunteers were randomly assigned to 1 of 2 experimental groups by using a computer-generated sequence of 139 nonunique, unsorted numbers with a range from 1 to 2 representing the groups. The main investigator (NV) kept the allocation sequence confidential and assigned the women to one or the other group.

Participants received 12 wk of supplementation with 900 mg/d of oral magnesium oxide corresponding to 300 mg bioavailable magnesium [Easymag (Sanofi-Aventis); treatment group] or no treatment (control group). This dose of magnesium was chosen because it corresponds to 94% of the RDA (19), and a once-daily administration was adopted to improve the participants’ compliance with the supplementation. The supplement was distributed to

![Figure 1](image-url)

**FIGURE 1.** Consolidated Standards of Reporting Trials diagram showing sample sizes at each stage of the study. 25(OH)D, 25-hydroxyvitamin D.
participants in sachets (daily doses of supplement) contained in cartons (monthly supplies). Biweekly phone calls were made by the research coordinator (LB) to record compliance with the treatment and any adverse events and to monitor whether participants were attending their fitness program. Their attendance was also recorded weekly in registers kept at the gyms.

At baseline, a trained dietitian asked the control group to avoid modifying their usual diet, and they received the same follow-up phone calls as the treated group. The 2 groups were followed up by 2 independent teams of physicians who conducted physical and body-composition tests at baseline and again after 12 wk. The same professional on each team performed the same test at both time points. To ensure the success of the masking procedure, these 2 teams of physicians did not know whether a given participant belonged to the treatment or the control group (the participants had been trained not to reveal this information to the personnel performing the tests).

**Outcomes**

All tests were performed at baseline and again after 12 wk. Our primary outcome was a change in the Short Physical Performance Battery (SPPB) (20):

1. The SPPB consists of 3 objective physical function tests: 4-m gait speed, repeated chair stands, and standing balance in increasingly challenging positions. Walking speed was calculated as the best performance achieved in 2 walks at the participant’s usual pace along a corridor 4 m long. For the chair stands test, the participants were asked to rise 5 times from a seated position as quickly as possible with their hands folded across their chest. For the standing balance tests, participants were asked to stand in 3 increasingly difficult positions (with their feet side by side and in semitandem and full-tandem positions) for 10 s each. Each test was scored from 0 (worst) to 4 (best), and the scores from all 3 tests were combined to obtain a composite score of 0 to 12—higher scores reflect better physical function.

Our secondary outcomes were variables of muscle strength:

1. Isometric knee extension torque and isokinetic (flexion and extension) strength were tested on the dominant side by using the dynamometer chair (Easytech s.r.l.). The participants were positioned upright with straps to fix their hips to the chair. For each of the 3 measurements, participants were asked to reach their maximal voluntary contraction. Three to 5 s after reaching their maximum effort, they were asked to stop the contraction. Each measurement was repeated 3 times, and patients rested for 2 to 3 min between trials. The highest-peak torque (PT) was used for the analysis.

2. Handgrip strength on the dominant side was measured by using DynEx electronic hand dynamometers. The participants were seated in a standard armchair with their shoulder adducted and neutrally rotated, their elbow flexed at 90°, and their forearm and wrist in a natural position. They were asked to grip the dynamometer smoothly, progressing up to their maximal strength in response to a voice command, without any wrenching or jerking motion. Three measurements were obtained on the dominant side with a 1-min rest between trials, and the highest measurement was used in our analyses.

We considered the following tertiary outcomes:

1. Blood and urine tests: venous blood samples were collected after the subjects fasted overnight. Biohumoral tests were completed at our Department of Laboratory Medicine, which belongs to the International Federation of Clinical Chemistry and Laboratory Medicine. Magnesium was measured in serum and in 24-h urine samples by using a standard colorimetric enzymatic method that consists of a coupled enzyme assay, which resulted in a colorimetric (450 nm) product proportional to the magnesium content. The intra- and interassay CVs for serum magnesium were 1.8% and 1.4%, respectively, at a concentration of 0.81 mmol/L, whereas the corresponding values for 24-h urinary magnesium were 0.6% and 1.6% at a concentration of 2.45 mmol/L. On the basis of quality-control materials (Liquichek Unassayed Chemistry Control; Bio-Rad), the interassay CVs for magnesium covered a range of concentrations from 1.8% to 5.1%. Serum 25(OH)D concentrations were measured by radioimmunoassay (DiaSorin); the intraassay and interassay CVs for 25(OH)D were 7.9% and 8.2%, respectively. Serum intact parathormone concentrations were measured by using a 2-step immunoradiometric assay (PTH-S; N-tact PTH SP; DiaSorin): the intraassay and interassay CVs for parathormone were 3.0% and 5.5%, respectively. For both procedures, the standards, controls, and samples were all analyzed in duplicate, and the samples were reanalyzed if the CVs of the duplicates were >10%.

2. Body composition: body weight and height were measured by trained staff. Body composition was assessed by dual-energy X-ray absorptiometry with a fan-beam technology (Hologic Discovery A) (21). The Appendicular Skeletal Muscle Mass Index, ie, the ratio of appendicular skeletal muscle mass to height (in kg/m²), was considered to be the best indicator of functional muscle mass in elderly people (22, 23).

3. Physical and dietary assessment: physical activity was measured by using the Physical Activity Scale for the Elderly (PASE), which is designed to assess physical activities in elderly adults within a 1-wk time frame (24). The scale covers 12 different activities, such as walking, sports, and housework, and is scored from 0 to 400 and more (no maximum score has been defined). The type of activity and its frequency, and the time spent on it, were recorded and the PASE scores were calculated as explained in the PASE manual (25). At baseline, a trained dietitian conducted a dietary assessment using a modified dietary assessment method involving an estimated 3-d record and a questionnaire on the frequency with which participants usually ate certain foods; data from the previous month were used as reference (26, 27). The usual food intake was converted into macronutrients and micronutrients by using a national food-composition table (28).

**Sample size and statistical analyses**

The required sample size was calculated from the difference of 0.28 points in total SPPB scores between the treated and control groups after 12 wk (29). With an SD of 0.5 points, the number of participants estimated to be needed in each group was 50 for an 80% power and α = 0.05. This sample size was also able to identify a difference of 4.0 ± 5.6 kg in handgrip strength as
significant, which had been found to be clinically significant in other studies (this applied to n = 31 participants per group) (30). Finally, because no unequivocal data are available for changes in isometric and isokinetic strength in old age, for the purposes of this study we assumed that a difference of 5 ± 5% for each variable was clinically significant (which applied to n = 16 participants per group).

Baseline characteristics were compared between the treatment and control groups by using independent t tests, chi-square tests, or Fisher’s exact test, as appropriate. Paired t tests were used for within-group comparisons of baseline and 12-wk data, and changes were calculated as the difference between the 2 values (Δ). Comparisons between data for the groups at 12 wk were computed by using a generalized linear model, adjusted for the baseline value of the corresponding test. In the ancillary analysis, we compared the changes in SPPB scores at 12 wk using a stratification for baseline SPPB total score and dietary magnesium intake. For interaction was calculated by using a generalized linear model with the baseline SPPB score categorized as ≤10 or 10 and the dietary magnesium intake as below or reaching the corresponding RDA. Pearson’s correlations were run on the combined data from both groups and were used to identify associations between serum indicators and primary outcome items. Significance was accepted if P ≤ 0.05, and all tests were 2-tailed. All analyses were performed by using SPSS 21.0 for Windows (SPSS Inc).

RESULTS

Participants

The mean (±SD) age of the sample as a whole was 71.5 ± 5.2 y, the mean weight was 66.3 ± 10.7 kg, and the mean BMI (in kg/m²) was 27.40 ± 4.18. During the follow-up, 10 participants were lost: these women were similar to the participants included in the analysis in terms of age, baseline physical performance and strength, blood and serum tests, body composition, and levels of physical activity (details not shown).

After the treated participants were separated from the control subjects, no significant differences emerged for age (71.8 ± 5.0 compared with 71.3 ± 5.4 y, respectively; P = 0.62), daily calorie intake (1454.76 ± 423.34 compared with 1526.57 ± 333.56 kcal; P = 0.29), or baseline magnesium intake (394.00 ± 230.13 compared with 372.05 ± 163.07 mg/d; P = 0.56). There were 24 participants with a magnesium intake below the RDA in the treated group and 30 in the control group (P = 0.86). The number of participants in the 2 groups taking vitamin D supplementation was also comparable: 23 in the treated group and 35 in the control group (P = 0.59).

Compliance

On the basis of sachet and carton counts and specific questions posed during the study and at the follow-up visit, the percentage of prescribed magnesium doses ingested by the treatment group was 90 ± 5.0%.

Primary outcome

**SPPB**

As shown in Table 1, no differences in total SPPB, single item scores, gait speed, or chair stand times were found between the 2 groups at baseline. When the variations in SPPB outcomes between the 2 groups were compared, the treated group showed a significantly greater improvement than did the control group for the SPPB walking item (Δ = 0.34 ± 0.10 points; P = 0.01),

![Table 1: Mean baseline characteristics and outcomes at follow-up, by group](image-url)

1 All values are means ± SDs. *P < 0.05. **P < 0.001. ***P < 0.0001. ASMMI, Appendicular Skeletal Muscular Mass Index; PASE, Physical Activity Scale for the Elderly; PT, peak torque; SPPB, Short Physical Performance Battery; 25(OH)D, 25-hydroxyvitamin D.

2 There were no statistically significant differences between the groups in any baseline test.

3 Changes were calculated as the differences between data at baseline and at follow-up after 12 wk (within-group comparison) by using paired t tests.

4 A generalized linear model was used for a between-groups comparison of the data obtained after 12 wk, adjusted for the baseline value of the corresponding test.
the total SPPB score ($\Delta = 0.41 \pm 0.24$ points; $P = 0.03$), the chair stands times ($\Delta = -1.31 \pm 0.33$ s; $P < 0.0001$), and walking speed ($\Delta = 0.14 \pm 0.03$ m/s; $P = 0.0006$) (Table 1).

**Ancillary analyses**

Among other factors that might have contributed to our findings, baseline total SPPB scores and dietary magnesium intake seem to be relevant. When our participants were stratified by median baseline total SPPB scores, we found that the items in the SPPB that improved significantly in the sample as a whole remained the same, irrespective of this distinction (Table 2). Dividing the sample between those who reached the RDA of dietary magnesium and those who did not, we found similar differences between the treated and control groups in terms of the changes in their chair stands time and walking speed (Table 3). On the other hand, the SPPB total score was significantly higher in the treated than in the control group, but only for participants with a magnesium intake below the RDA, and their better SPPB score was probably the result of a significant improvement in the SPPB walking item.

**Secondary outcome: PT isometric and isokinetic leg and handgrip strength measurements**

At baseline, no differences in PT leg or handgrip strength were found between the groups. After 12 wk, no significant within- or between-group changes were found in PT isometric and isokinetic strength. The changes in handgrip strength were also similar between the groups (Table 1).

**Tertiary outcomes**

**Blood and urine tests**

No significant differences in serum and 24-h urinary magnesium were found at baseline. A comparison of the changes for these 2 outcomes between the groups showed that both serum ($\Delta = 0.02 \pm 0.007$ mmol/L; $P = 0.03$) and urinary magnesium ($\Delta = 1.45 \pm 0.31$ mmol/24 h; $P < 0.0001$) were significantly higher in the treated than in the control group. The changes from baseline in 25(OH)D and parathormone did not differ significantly between the 2 groups (Table 1).

**Body composition**

When the Appendicular Skeletal Muscle Mass Index was used as an indicator of functional lean mass, no difference was found between the groups at baseline, and no significant differences were found between the groups at 12 wk (Table 1). Similarly, no significant differences in weight, height, BMI or total fat mass, and fat-free mass emerged between the groups at baseline, and the situation remained the same after 12 wk (details not shown).

**Physical assessment**

No significant between-groups differences were found in the levels of physical activity, based on the PASE score, were found at baseline ($P = 0.68$) or at the follow-up ($P = 0.67$) visits (Table 1). Finally, the number of participants who attended $\geq 80\%$ of their fitness program meetings was high and was similar between the 2 groups (45 in the treated and 60 in the control group; $P = 0.78$).

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**Table 2** Primary outcome in the treated and control groups by baseline SPPB score

<table>
<thead>
<tr>
<th>Measure</th>
<th>Baseline</th>
<th>12 wk</th>
<th>Change</th>
<th>Baseline</th>
<th>12 wk</th>
<th>Change</th>
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<th>Baseline</th>
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<th>Change</th>
<th>Baseline</th>
<th>12 wk</th>
<th>Change</th>
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<tbody>
<tr>
<td>SPPB-balance</td>
<td>0.63</td>
<td>0.78</td>
<td>0.15</td>
<td>0.60</td>
<td>0.65</td>
<td>0.05</td>
<td>0.60</td>
<td>0.65</td>
<td>0.05</td>
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<tr>
<td>SPPB-30</td>
<td>0.79</td>
<td>0.86</td>
<td>0.07</td>
<td>0.70</td>
<td>0.74</td>
<td>0.04</td>
<td>0.70</td>
<td>0.74</td>
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<tr>
<td>SPPB-chair</td>
<td>0.50</td>
<td>0.58</td>
<td>0.08</td>
<td>0.43</td>
<td>0.51</td>
<td>0.08</td>
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<tr>
<td>Chair stands times (s)</td>
<td>2.10</td>
<td>1.95</td>
<td>0.15</td>
<td>2.12</td>
<td>1.95</td>
<td>0.17</td>
<td>2.12</td>
<td>1.95</td>
<td>0.17</td>
<td>2.12</td>
<td>1.95</td>
<td>0.17</td>
<td>2.12</td>
<td>1.95</td>
<td>0.17</td>
<td>2.12</td>
<td>1.95</td>
<td>0.17</td>
<td></td>
<td></td>
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<tr>
<td>Walking speed (m/s)</td>
<td>0.16</td>
<td>0.14</td>
<td>0.02</td>
<td>0.16</td>
<td>0.14</td>
<td>0.02</td>
<td>0.16</td>
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All values are means $\pm$ SDs. *P* < 0.05, **P** < 0.01, ***P*** < 0.001. SPPB, Short Physical Performance Battery.
Correlations

Pearson’s correlations were used to determine whether the changes in serum or urinary magnesium were associated with changes in primary outcomes. Increases in serum magnesium were significantly associated with improvements in gait speed ($r = 0.20$, $P = 0.03$) and chair stand times ($r = -0.18$, $P = 0.05$) (Figure 2, A and B). No other significant correlations emerged between serum magnesium supplementation and the other physical performance tests considered. Changes in 24-h urinary magnesium did not correlate with the results of the physical performance tests.

Adverse events

No severe adverse events were reported in either group. Mild adverse effects of magnesium supplementation included one case of diarrhea and 2 of itching, which both regressed spontaneously after the treatment was suspended. Finally, no cases of hypermagnesemia were observed in the group receiving supplementation.

### DISCUSSION

In the current study, we aimed to investigate the effect of magnesium supplementation on physical performance and muscle strength in a sample of healthy, still physically active elderly women. Our main findings were that magnesium supplementation significantly improved the total SPPB score and walking speed and chair stand times. At the same time, the improvement in total SPPB score in the treated group’s was twice as high as that in the control group and high enough to be clinically relevant in terms of a meaningful change in physical performance proposed for older people (29, 31).

Magnesium supplementation positively affected gait speed and chair stand times. The improvement in gait speed was substantial: the treated group had a mean improvement of ~12 m/min versus the baseline. Like the improvement in total SPPB score, this finding is important in clinical terms, because gait speed is the only item in the SPPB that can be used as a single variable for diagnosing sarcopenia (a condition involving a degenerative loss of skeletal muscle mass), because it is an independent predictor of adverse health events (23). Finally, magnesium supplementation strongly improved chair stand time, which appears to be the most multidimensional item in the SPPB, including many features essential to elderly people’s wellness, such as lower limb strength, balance, and psychological aspects (32). On the other hand, we found no differences in the balance tests, probably attributable to a ceiling effect. The role of a ceiling effect in the SPPB test is further supported by the chair stands item: when it was considered as a score, we found no significant difference between the groups, but when it was considered as a continuous variable it showed the largest improvement among the SPPB items.

The effects of magnesium supplementation on the SPPB scores were more evident in participants with a dietary magnesium intake below the RDA, confirming previous reports that magnesium supplementation has beneficial effects on exercise performance in magnesium-deficient individuals (16). Although we only assessed dietary intake at baseline, any changes in diet were not assessed dietary intake at baseline, any changes in diet were
Magnesium and physical performance in the elderly.

May have had an age-related subclinical form of magnesium deficiency, so the increase in their serum magnesium concentration would reflect a positive effect in subjects with limited magnesium reserves. This assumption is supported by the fact that all our participants had normal serum magnesium concentrations at baseline, but about one-half of them had a low dietary magnesium intake, 6% had serum magnesium concentrations ≥0.75 mmol/L (39), and 5% had excreted magnesium concentrations <2.5 mmol/d—the lower limit of normality at our laboratory. Serum magnesium concentrations also proved clinically more significant than urinary magnesium: we only found a significant correlation between serum magnesium (but not urinary magnesium) and changes in gait speed and chair stand times, which suggests that this simple test could be useful as a first-line investigation when assessing physical performance in older people.

In addition to the fact that dietary intake was investigated only at baseline, the current study had some other limitations that need to be mentioned. First, it was not a double-blind trial, although only the research coordinator and main investigator knew whether participants were taking magnesium supplements or not, whereas the team members did not, and the outcomes were tested by using machines or standardized procedures. It is impossible to say how much of the differences found between the groups might have been attributable to the intervention group’s expectations. Moreover, this trial was conducted only in women; therefore, these findings cannot be generalized to men. On the other hand, a strength of our work was its use of a global assessment of physical performance and muscle strength, explored by using reliable and reproducible methods. To the best of our knowledge, this was the first trial to include only elderly people and to analyze the association of magnesium supplementation with physical performance, considering both upper and lower limb strength items. Another strength was the comprehensive assessment of magnesium status, which was investigated in terms of dietary intake, and serum and 24-h urinary magnesium concentrations. Finally, body composition was assessed by dual-energy X-ray absorptiometry, which is the preferred method for assessing body composition for research purposes and in the clinical setting (23).

In conclusion, oral supplementation with 300 mg Mg in the form of magnesium oxide for 12 wk had a significant positive effect on physical performance, as assessed with the SPPB and gait speed and chair stand times in healthy elderly women. These findings suggest a role for magnesium supplementation in preventing or delaying the age-related decline in physical performance, particularly in magnesium-deficient individuals. Further research is needed to understand the influence of magnesium supplementation on physical performance in elderly people with different magnesium concentrations.

Our sincerest gratitude goes to the gym coordinator, Flavio Martinello, for helping with recruitment. We thank the participants of the study, the nurses at the San Massimo Clinic (Padova Hospital, Laboratory Medicine), and Monica and Elisabetta (Sanofi-Aventis), who helped us to find the free samples given to the participants. Written permission was obtained from all persons named in this section.

The authors’ responsibilities were as follows—NV, AC, and GS: designed the trial; LB, SC, SP, GB, and FM: collected the data; EDT and EP: conducted the analysis and interpreted the data; NV, LB, FB, and MDR: wrote the manuscript; and EM and GS: took primary responsibility for the final content. None of the authors declared a conflict of interest.
REFERENCES


