Association of Serum Magnesium Levels With Risk of Intracranial Aneurysm

A Mendelian Randomization Study

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Abstract

Objective

Magnesium has been implicated in regulating blood pressure and vascular endothelial cell function, but its role in the pathophysiology of intracranial aneurysm is not known. Here we performed a Mendelian randomization analysis to investigate the association between serum magnesium concentration and risk of intracranial aneurysm.

Methods

Five single-nucleotide polymorphisms strongly associated with serum magnesium concentrations in a genome-wide association study in 23,829 individuals of European ancestry were used as genetic instruments. Genetic association estimates for intracranial aneurysm were obtained from a genome-wide association study in 79,429 individuals (7,495 cases and 71,934 controls). The inverse variance weighted method was used in the primary analyses to obtain the causal estimates.

Results

Higher genetically predicted serum magnesium concentrations were associated with lower risk of intracranial aneurysm. The odds ratios per 0.1 mmol/L increment in genetically predicted serum magnesium concentrations were 0.66 (95% confidence interval [CI] 0.49–0.91) for intracranial aneurysm (unruptured and ruptured combined), 0.57 (95% CI 0.30–1.06) for unruptured intracranial aneurysm, and 0.67 (95% CI 0.48–0.92) for aneurysmal subarachnoid hemorrhage.

Conclusion

This study provides evidence to support that increased serum magnesium concentrations reduce the risk of intracranial aneurysm and associated hemorrhage.

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Glossary

CI = confidence interval; **LD** = linkage disequilibrium; **MR** = mendelian randomization; **OR** = odds ratio; **SNP** = single nucleotide polymorphism.

Intracranial aneurysm rupture and resultant subarachnoid hemorrhage is associated with a high rate of morbidity and mortality.[1,2] Serum magnesium concentrations have been implicated in regulating blood pressure and function of the vascular endothelium.[3-5] However, whether increased serum magnesium concentrations affect the risk of intracranial aneurysm and related subarachnoid hemorrhage is not known.

To investigate this, we leveraged randomly allocated genetic variants related to serum magnesium concentrations as instrumental variables in a 2-sample mendelian randomization (MR) study assessing whether higher genetically predicted serum magnesium concentrations are associated with a reduced risk of intracranial aneurysm. A multivariable MR analysis was further performed to investigate whether any potential association may be mediated through effects on blood pressure.

Methods

Genetic Instruments and Data Sources

As instrumental variables for the primary analyses, we used single nucleotide polymorphisms (SNPs) associated with serum magnesium concentrations at genome-wide significance ($p < 5 \times 10^{-8}$) in a genome-wide association study in 23,829 individuals of European ancestry.[6] Six independent SNPs in different genetic loci and on different chromosomes were identified and proposed as instrumental variables for serum magnesium concentrations. In complementary analyses, we added 2 uncorrelated SNPs in or near known magnesium transport genes and associated with serum magnesium concentrations after applying a Bonferroni correction for the number of genetic variants examined in each region.[6] Summary statistics data for intracranial aneurysm in individuals of European ancestry were obtained from a genome-wide association study of 23 cohorts comprising 79,429 individuals (7,495 cases [69% with ruptured intracranial aneurysm, 28% with unruptured intracranial aneurysm, and 3.8% with unknown rupture status] and 71,934 controls).[7] Genetic association estimates for systolic blood pressure were taken from a genome-wide association study of 757,601 individuals of European ancestry.[8]

Standard Protocol Approvals, Registrations, and Patient Consents

Only summary-level data were analyzed in this study, for which appropriate ethical approval and participant consent had previously been acquired in the original genome-wide association studies. The analyses were approved by the Swedish Ethical Review Authority (2019–03986).

Statistical Analysis

In our main analyses, we applied the multiplicative random-effects inverse-variance weighted method to estimate the total association of serum magnesium concentrations with intracranial aneurysm.[9] To assess the potential presence of directional pleiotropy, MR-Egger regression analysis was performed.[9] Finally, to evaluate whether the results may be driven by potential outliers, we used the MR pleiotropy residual sum and outlier method.[9] To evaluate whether blood pressure may mediate any association, we implemented a multivariable MR model with adjustment for genetically predicted systolic blood pressure. The weighted median method was used as a complementary method.[9] We scaled all odds ratios (ORs) per 0.1 mmol/L increase in serum magnesium concentrations, which corresponds to an approximate 1 SD. Analyses were conducted using the mrrobust package[10] for Stata (StataCorp) and the MendelianRandomization package[11] for R.

Data Availability

All data generated or analyzed during this study are included in the table.

Results

Of the 6 SNPs associated with serum magnesium at the genome-wide significance threshold, 1 SNP was unavailable in the intracranial aneurysm datasets and no proxy SNP was available at a linkage disequilibrium (LD) $R^2 > 0.6$. The other 5 SNPs (including 1 proxy SNP in strong LD with the original SNP) were used as instrumental variables for serum magnesium in the primary analyses. For the 2 SNPs in or near magnesium transport genes, we used proxy SNPs in strong or complete LD with the original SNPs (table). Information on the SNPs used as instrumental variables and their associations with intracranial aneurysm is shown in the table.

Genetically predicted serum magnesium concentrations were inversely associated with risk of intracranial aneurysm. In the analyses based on the 5 genome-wide significant SNPs, the ORs per 0.1 mmol/L increase in genetically predicted magnesium concentrations were 0.66 (95% confidence interval [CI] 0.49–0.91) for intracranial aneurysm, 0.57 (95% CI 0.30–1.06) for unruptured intracranial aneurysm, and 0.67 (95% CI 0.48–0.92) for aneurysmal subarachnoid hemorrhage. The sensitivity analysis based on the weighted median method provided similar results (corresponding ORs 0.64 [95% CI 0.45–0.91], 0.62 [95% CI 0.32–1.20], and 0.66 [95% CI 0.44–0.98]). There was no evidence of directional pleiotropy (all p values for the MR-Egger intercept >0.60), and no outlier was found in the MR pleiotropy residual sum and outlier method.

Table Characteristics of the Single Nucleotide Polymorphisms Used as Instrumental Variables for Serum Magnesium Concentrations and Their Associations With Intracranial Aneurysm

rsID	Chr	Gene	EA	Serum magnesium concentrations β (SE)[a]	p Value	IA (unruptured and ruptured) β (SE)	p Value	Unruptured IA β (SE)	p Value	Ruptured IA (aSAH) β (SE)	p Value
rs4072037	1	MUC1	T	0.010 (0.001)	2.01E-36	−0.038 (0.021)	0.07	−0.048 (0.039)	0.22	−0.036 (0.024)	0.14
rs7965584[b]	12	ATP2B1	A	0.007 (0.001)	1.05E-16	−0.050 (0.022)	0.02	−0.098 (0.038)	0.01	−0.036 (0.027)	0.18
rs3925584	11	DCDC5	T	0.006 (0.001)	5.20E-16	0.016 (0.022)	0.48	0.029 (0.039)	0.46	0.008 (0.026)	0.75
rs13146355	4	SHROOM3	A	0.005 (0.001)	6.27E-13	−0.026 (0.019)	0.17	−0.010 (0.034)	0.77	−0.044 (0.023)	0.05
rs448378	3	MDS1	A	0.004 (0.001)	1.25E-8	−0.036 (0.021)	0.09	−0.062 (0.038)	0.11	−0.024 (0.024)	0.32
rs3740393[c]	10	CNNM2	C	0.006 (0.001)	8.58E-7	−0.174 (0.030)	1.11E-8	−0.17 (0.056)	0.002	−0.018 (0.035)	3.98E-7
rs6746896[c]	2	CNNM4	G	0.004 (0.001)	7.01E-5	−0.004 (0.023)	0.87	0.041 (0.041)	0.32	−0.023 (0.027)	0.41

Abbreviations: aSAH = aneurysmal subarachnoid hemorrhage; Chr = chromosome; EA = effect allele (the allele that associates with higher serum magnesium concentrations); IA = intracranial aneurysm.
[a] The β coefficients represent the increase in serum magnesium concentrations in mmol/L per additional effect allele.
[b] The single nucleotide polymorphism was unavailable in the intracranial aneurysm datasets but a proxy genetic variant (rs10858938, effect allele = G) in linkage disequilibrium (r^2 = 0.96 in European populations) was identified and replaced this genetic variant.
[c] Used in complementary analyses that included genetic variants in or near known magnesium transport genes and associated with serum magnesium after applying a Bonferroni correction for the number of genetic variants assessed in each region.[6] The single nucleotide polymorphisms described in the original study[6] were unavailable in the intracranial aneurysm datasets, so proxy genetic variants in linkage disequilibrium (r^2 > 0.9) were used: rs72841270 (effect allele = G) for rs3740393 and rs56351161 (effect allele = A) for rs6746896.
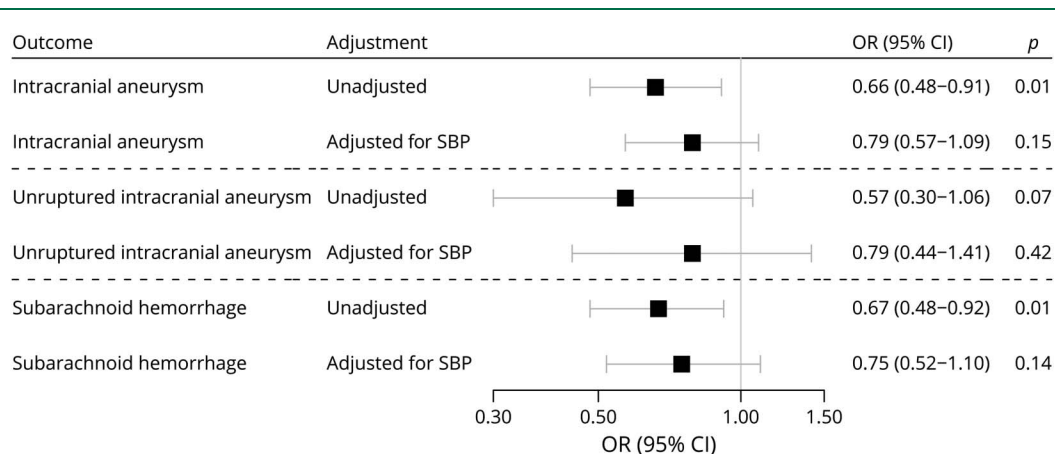
Adjustment for genetically predicted systolic blood pressure through a multivariable MR model attenuated the associations of genetically predicted serum magnesium with all 3 considered outcomes, consistent with a partial mediating effect (figure).

The complementary analyses incorporating 2 SNPs in or near magnesium transport genes yielded similar results. In these analyses based on 7 SNPs, the ORs per 0.1 mmol/L increase in genetically predicted magnesium concentrations were 0.55 (95% CI 0.32–1.00) for intracranial aneurysm, 0.53 (95% CI 0.25–1.10) for unruptured intracranial aneurysm, and 0.55 (95% CI 0.32–0.97) for aneurysmal subarachnoid hemorrhage.

Discussion

This MR study provides evidence to support that higher serum magnesium concentrations reduce the risk of intracranial aneurysm and aneurysmal subarachnoid hemorrhage, with systolic blood pressure mediating part of this effect. Whereas magnesium supplementation has been demonstrated to increase serum magnesium concentrations and reduce blood pressure in randomized clinical trials,[3] to our knowledge this is the first MR study to identify a potential causal association between serum magnesium concentrations and risk of intracranial aneurysm and aneurysmal subarachnoid hemorrhage. The associations of genetically predicted serum

Figure Association Between Genetically Predicted Serum Magnesium Concentrations and Intracranial Aneurysm

Outcome	Adjustment	OR (95% CI)	p
Intracranial aneurysm	Unadjusted	0.66 (0.48–0.91)	0.01
Intracranial aneurysm	Adjusted for SBP	0.79 (0.57–1.09)	0.15
Unruptured intracranial aneurysm	Unadjusted	0.57 (0.30–1.06)	0.07
Unruptured intracranial aneurysm	Adjusted for SBP	0.79 (0.44–1.41)	0.42
Subarachnoid hemorrhage	Unadjusted	0.67 (0.48–0.92)	0.01
Subarachnoid hemorrhage	Adjusted for SBP	0.75 (0.52–1.10)	0.14

Estimates are scaled per genetically predicted 0.1 mmol/L (about 1 SD) increase in serum magnesium concentrations. Adjustment for genetically predicted systolic blood pressure (SBP) was made using a multivariable mendelian randomization model. CI = confidence interval; OR = odds ratio.

magnesium with risk of intracranial aneurysm and aneurysmal subarachnoid hemorrhage did not fully attenuate after adjustment for genetically predicted systolic blood pressure, suggesting that magnesium may also affect the risk of these outcomes via other mechanisms. In addition to a blood pressure–lowering effect, increased magnesium concentrations may reduce the risk of intracranial aneurysm rupture by improving endothelial function[4,5] and reducing oxidative stress.[12]

A strength of this study is the MR design, which is less prone to confounding compared with conventional observational studies. Other major strengths are the relatively large number of cases of intracranial aneurysm and the robustness of the findings in different MR sensitivity analyses that are more robust to the inclusion of pleiotropic variants. Population stratification bias was minimized because all analyses were restricted to populations of European ancestry and adjustment was made for principal components for ancestry in the original genome-wide association studies from which summary data were used. In terms of limitations, we cannot rule out that our genetic proxies for serum magnesium affect intracranial aneurysm risk through alternative pathways that may violate the requisite assumptions of MR. Another limitation is that we did not have access to appropriate data for a bidirectional MR analysis to investigate whether biological mechanisms predisposing to intracranial aneurysm influence circulating magnesium levels. Finally, the genetic variants used to proxy the effect of modifying serum magnesium concentration reflect small, lifelong effects in serum magnesium concentrations, while in contrast a clinical intervention would typically exert a greater change in serum magnesium concentrations later in life. Caution should be taken when extrapolating findings from MR to infer the effect of a clinical intervention, and clinical trials are warranted to guide optimal practice.

This MR study provides evidence to support that higher serum magnesium concentrations reduce the risk of intracranial aneurysm and aneurysmal subarachnoid hemorrhage. These findings add to the growing body of evidence highlighting a beneficial role of higher magnesium for preventing cerebrovascular and cardiovascular diseases.[13-15]

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Study Funding

Disclosure

Publication History

Appendix Authors

Name	Location	Contribution
Susanna C. Larsson, PhD	Uppsala University, Sweden	Designed and conceptualized study, analyzed and interpreted the data, drafted the manuscript for intellectual content
Dipender Gill, BMBCh, PhD	Imperial College London, UK	Designed and conceptualized study, interpreted the data, revised the manuscript for intellectual content

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