INTRODUCTION

Vitamin D is an endogenously synthesized hormone which plays multiple roles in the immune and endocrine systems, in addition to its role in regulating mineral metabolism in the skeletal system. Many studies have been conducted to understand its effects on atherosclerosis in coronary and cerebrovascular diseases. As an active metabolite, 25-hydroxy vitamin D (25(OH)D) originally exerts a protective effect on vascular endothelial stability. However, both vitamin D deficiency and excess can exacerbate atherosclerosis via various molecular mechanisms.

Through decades of trials, carotid artery stenting (CAS) has been recognized as a reasonable treatment option in carotid stenosis compared with carotid endarterectomy. Since residual stenosis impedes favorable periprocedural outcomes and long-term efficacy, ensuring sufficient balloon angioplasty and stent expansion is of great importance to neurointerventionists. Considering the role of vitamin D in the development of atherosclerosis, we examined whether serum 25(OH)D levels are associated with the expansion rate of carotid stenting.

Received: November 9, 2021
Revised: December 6, 2021
Accepted: December 26, 2021

Correspondence:
Chi Kyung Kim, MD, PhD
Department of Neurology, Korea University Guro Hospital, Korea University College of Medicine, Seoul, Korea
Tel: +82-2-2626-1250
Fax: +82-2-2626-2249
E-mail: ckkim7@gmail.com

The two authors contributed equally to the work.
D level influences the expansion rate of the carotid stent.

SUBJECTS AND METHODS

1. Patient and data collection

From May 2018 to June 2021, data on 78 cases of elective cervical CAS performed in Korea University Guro Hospital were retrospectively collected from the Korea University Stroke Registry (KUSR). The KUSR has been used in several research papers published in authorized journals. Among 78 cases, 32 patients with absent laboratory results were excluded. Laboratory tests included low-density lipoprotein (LDL) cholesterol, hemoglobin A1c, creatinine, C-reactive protein (CRP), D-dimer, total serum calcium, serum phosphate, parathyroid hormone, and 25(OH)D. All laboratory tests were performed within a month of stent insertion. Data on balloon diameter, number of angioplasties, types of stents, and stent diameter were collected. The balloon diameter was averaged for the multiple angioplasty cases. Stent types were divided into closed and open-cell stents. The stent diameter was averaged for the tapered stent cases.

In Korea University Guro Hospital, elective cervical CAS is suggested when the patient has stenosis of 70% or more, or an ulcerative plaque as indicated in computed tomography angiography (CTA), magnetic resonance angiography (MRA) or digital subtraction angiography (DSA); stenosis is determined as being symptomatic, and aspirin 100 mg or more and clopidogrel 75 mg or more is administered at least 7 days before CAS. The procedures for our cases were conducted when all indications were fulfilled and performed by a neuro-interventionist with over 15 years of experience. All CAS procedures were performed after obtaining informed consent.

2. Definitions and interpretation

The investigators defined a case as symptomatic when transient ischemic attack (TIA), monocular blindness, and cerebral infarction occurred ipsilateral to the stenosed target carotid artery, and assessment of relevance between carotid stenosis and symptoms was conducted in the Department of Neurology. A low serum level of 25(OH)D was specified by levels under 15.1 ng/mL. The cervical carotid artery was specified as the region of the artery running from the common carotid artery to the cervical portion of the internal carotid artery as seen in CTA, MRA, or DSA. The stenosis degree before and after stent insertion was calculated as the ratio of the diameter of the plaque measured at the most stenosed point to the distal normal luminal diameter, which is the North American Symptomatic Carotid Endarterectomy Trial (NASCET) method, in DSA. We defined the stent expansion rate using the following equation: \( \frac{1 - \text{post-stent stenosis degree}}{\text{pre-stent stenosis degree}} \times 100 \% \). Successful stent expansion was specified as a stent expansion rate of 70% or more in DSA. All image assessments were performed by two neurointerventionists (J.H., H.J.) with substantial consistency (Cohen’s \( \kappa = 0.85 \)).

3. Statistical analysis

Baseline demographic, laboratory, and imaging data of the two groups were compared according to serum 25(OH)D levels, above or below 15.1 ng/mL. To search for independent factors for successful stent expansion, multivariable logistic regression analysis was conducted with variables that were assumed to be clinically relevant by investigators, or which had a \( p \)-value of less than 0.1 in comparison to baseline characteristics. Age, smoking status, hypertension, dyslipidemia, and diabetes mellitus, which are regarded as traditional risk factors for carotid stenosis, as well as the initial carotid stenosis degree, were included in the regression analysis according to the researchers’ agreement. Male sex and the serum levels of D-dimer, CRP, total calcium, phosphate, and parathyroid hormone were included in the regression analysis according to previous studies. Continuous variables were analyzed using Student’s \( t \)-test and Mann–Whitney test, whereas the frequencies of categorical variables were compared using the \( \chi^2 \) test and Fisher’s exact test. Spearman’s correlation coefficient was used to determine the linear correlation between serum vitamin D level and stent expansion rate. Statistical significance was set at \( p < 0.05 \).
USA) was used.

**RESULTS**

The investigators assessed the retrospective data of 46 patients. The median age was 75 years (interquartile range [IQR] 69-79), and five men (10.9%) were included. Among the 46 patients, 25 (54.3%) had low serum 25(OH)D level. Age and LDL-cholesterol levels were analyzed using the Mann–Whitney test, and the frequency of sex and smoking status were assessed using Fisher’s exact test. There was no significant difference in the baseline demographic data and pre-stent stenosis degree between the groups. The serum calcium and phosphate levels did not differ between the two groups. Of the laboratory results, the level of parathyroid hormone was significantly higher in the low 25(OH)D group (37.6±3.6 vs. 27.0±3.1, respectively; p=0.03). The successful stent expansion rate was significantly higher in the low 25(OH)D level group (48.0% vs. 19.0%, respectively; p=0.04) (Table 1). The diameter of the balloon, number of angioplasties, types of stents, and stent diameters did not differ between the groups. Serum 25(OH)D levels were negatively correlated with stent expansion rate (Spearman’s correlation coefficient =-0.34, p=0.02) (Fig. 1).

Logistic regression analysis was performed to identify independent factors associated with successful stent ex-

---

**Table 1. Baseline characteristics of patients**

|                      | 25-OH vitamin D <15.1 ng/mL (n=25, 54.3%) | 25-OH vitamin D ≥15.1 ng/mL (n=21, 45.7%) | p-value |
|----------------------|------------------------------------------|------------------------------------------|---------|
| Median age, years    | 73 (68-80)                               | 76 (70-80)                               | 0.63    |
| Male sex             | 4 (16.0)                                 | 1 (4.8)                                  | 0.36    |
| Hypertension, yes    | 12 (57.1)                                | 20 (80.0)                                | 0.09    |
| Diabetes mellitus, yes | 9 (36.0)                                 | 7 (33.3)                                 | 0.85    |
| Dyslipidemia, yes    | 8 (32.0)                                 | 5 (23.8)                                 | 0.54    |
| Ever smoking, yes    | 3 (12.0)                                 | 2 (9.5)                                  | 1.00    |
| LDL-cholesterol, mg/dL | 83.0 (69.0-110.5)                        | 72.0 (50.0-99.5)                         | 0.21    |
| Creatinine, mg/dL    | 0.87±0.04                                | 0.91±0.05                                | 0.52    |
| C-reactive protein, mg/L | 3.76±1.23                             | 2.44±0.59                                | 0.37    |
| D-dimer, μg/mL       | 1.36±0.34                                | 0.86±0.13                                | 0.21    |
| Total calcium, mg/dL | 8.45±0.24                                | 8.75±0.07                                | 0.27    |
| Phosphate, mg/dL     | 3.17±0.16                                | 3.33±0.13                                | 0.44    |
| Parathyroid hormone, pg/mL | 37.60±3.58                        | 27.00±3.14                               | 0.03    |
| Balloon angioplasty  |                                          |                                          |         |
| Balloon diameter, mm | 3.91±0.64                                | 3.94±0.62                                | 0.87    |
| Number of angioplasty, times | 4                                      | 4                                        | 1.00    |
| Stent                |                                          |                                          |         |
| Types of stent, open-cell | 15 (71.4)                      | 18 (72.0)                                | 0.97    |
| Stent diameter, mm   | 7.74±0.94                               | 8.04±0.91                               | 0.27    |
| Pre-stent stenosis degree, NASCET, % | 78.50±13.8             | 77.40±11.1                               | 0.56    |
| Stent expansion rate, % | 63.9±17.3                      | 57.0±18.1                                | 0.19    |
| Successful stent expansion rate | 12 (48.0)                      | 4 (19.0)                                | 0.04    |

Values are presented as number (%), median (interquartile range), or mean±standard deviation. Stenosis degree was measured by NASCET criteria. Successful stent expansion was defined as a 70% or more increase of arterial lumen compared to before and afterward the procedure. 25-OH; vitamin D indicates 25-hydroxy vitamin D, LDL; low-density lipoprotein, NASCET; North American Symptomatic Carotid Endarterectomy Trial.
pansion. Low serum 25(OH)D level was significantly re-
lated (odds ratio [OR], 3.92; 95% confidence interval [CI], 1.03-15.0), whereas parathyroid hormone level was not 
associated (OR, 1.01; 95% CI, 0.97-1.05) with successful 
stent expansion in univariate analysis. In multivariable 
logistic regression analysis, age, male sex, hypertension, 
dyslipidemia, ever smoking status, and low 25(OH) 
D level, pre-stent stenosis degree, and serum levels of 
CRP, D-dimer, total calcium, phosphate, and parathy-
roid hormone were included as possible covariates; low 
serum 25(OH)D level was independently associated with 
successful stent expansion rate (adjusted OR, 6.42; 95% 
CI, 1.09-37.7) (Table 2).

Table 2. Logistic regression analysis for successful stent expansion

|                          | Crude odd ratio | p-value | Adjusted odd ratio | p-value |
|--------------------------|-----------------|---------|--------------------|---------|
| Age, years               | 1.03 (0.94-1.13) | 0.514   | 1.11 (0.96-1.28)   | 0.164   |
| Male sex, yes            | 0.43 (0.04-4.24) | 0.473   | 0.11 (0.01-2.25)   | 0.150   |
| Hypertension, yes        | 0.94 (0.25-3.51) | 0.930   | 0.48 (0.06-3.68)   | 0.482   |
| Diabetes mellitus, yes   | 0.79 (0.22-2.86) | 0.714   | 1.12 (0.17-7.51)   | 0.904   |
| Dyslipidemia, yes        | 1.25 (0.33-4.73) | 0.743   | 1.16 (0.16-8.28)   | 0.885   |
| Ever smoking, yes        | 3.23 (0.48-21.7) | 0.228   | 4.30 (0.21-89.0)   | 0.345   |
| C-reactive protein, mg/L | 0.93 (0.78-1.10) | 0.714   | 0.92 (0.73-1.16)   | 0.476   |
| D-dimer, μg/mL           | 1.04 (0.66-1.63) | 0.872   | 1.23 (0.44-3.72)   | 0.794   |
| Pre-stent stenosis degree, % | 0.96 (0.87-1.07) | 0.445   | 0.94 (0.81-1.09)   | 0.381   |
| Total calcium, mg/dL     | 1.09 (0.54-2.23) | 0.811   | 1.15 (0.21-6.46)   | 0.872   |
| Phosphate, mg/dL         | 1.02 (0.43-2.42) | 0.966   | 1.40 (0.20-9.65)   | 0.736   |
| Parathyroid hormone, pg/mL | 1.01 (0.97-1.05) | 0.603   | 1.03 (0.96-1.10)   | 0.437   |
| Low 25-OH vitamin D, yes | 3.92 (1.03-15.0) | 0.046   | 6.42 (1.09-37.7)   | 0.039   |

FIG. 1. Correlation analysis using Spearman’s correlation coefficient. The level of serum vitamin D and stent expansion rate were negatively correlated.

FIG. 2. Two representative cases with similar pre-stenting stenosis degrees. Anterior-posterior (AP) and lateral views of the angiogram pre- and post-stenting are demonstrated. Case 1) A patient with a successful expansion rate (90.3%), serum 25-OH vitamin D level below 15.1 ng/mL (14.0 ng/mL), and a pre-stent stenosis degree of 76.1%. (A) Pre-stenting AP and lateral views and (B) post-stenting AP and lateral views in the angiogram, respectively. Case 2) A patient without a successful expansion rate (23.5%), serum 25-OH vitamin D level above 15.1 ng/mL (27.1 ng/mL), and a pre-stent stenosis degree of 76.9%. (C) Pre-stenting AP and lateral views and (D) post-stenting AP and lateral views in the angiogram, respectively.
Two representative cases, compatible with our study results, are shown in Fig. 2. Case 1 demonstrates a patient with a successful stent expansion rate (90.3%), serum 25(OH)D level below 15.1 ng/mL (14.0 ng/mL), and a pre-stent stenosis degree of 76.1% (Fig. 2A, B). Case 2 demonstrates a patient without a successful stent expansion rate (23.5%), serum 25(OH)D level above 15.1 ng/mL (27.1 ng/mL), and a pre-stent stenosis degree of 76.9% (Fig. 2C, D).

DISCUSSION

In summary, among the baseline characteristics, the rate of successful stent expansion showed a significant difference between the low- and high-vitamin D groups. In the multivariate logistic regression analysis, low vitamin D levels were independently associated with successful stent expansion.

In the past 20 years, plaque composition has emerged as a major influential factor in determining whether coronary or carotid stenosis exerts a hazardous effect on disease prognosis. The histologic features of unstable plaques include intraplaque hemorrhage, a lipid-rich necrotic core, and a thin fibrous cap. In contrast to the latter, mature stage of atherosclerosis with more fibrotic, calcified lesions in “soft” plaques are related to early stage atherosclerosis. With larger neointimal and plaque areas, unstable plaques are susceptible to arterial remodeling, and are associated with in-stent restenosis, plaque fragmentation, and embolism.

In this manner, unstable carotid plaques increase the ipsilateral stroke by 4- to 12-times independent of the degree of stenosis.

A recent study revealed that low vitamin D levels make carotid plaques vulnerable by inducing intraplaque hemorrhage. At low vitamin D levels, the local angiotensin system loses inhibition and becomes abnormally activated, resulting in microvascular leakage and inflammation. However, the mechanically soft nature of unstable plaques was related to greater luminal gain after coronary stenting than calcified, stable plaques. In this context, the plaque characteristics in the low vitamin D group of our study could be more vulnerable and soft, thus exhibiting more successful stent expansion than in the high vitamin D group.

Although reducing the degree of residual stenosis is crucial for the prognosis of CAS, the possibility of an underlying harmful pathology should not be neglected. Conversely, high vitamin D levels are associated with vascular calcification by promoting vascular osteogenesis and mineralization. Since calcification reduces vascular stretch capacity and decreases luminal gain after CAS, the high vitamin D group in our study might have less successful stent expansion.

Our study had several limitations. First, this was a single-center cross-sectional study with a limited sample size. However, most cases were consecutively collected with the preinvestigated vitamin D levels. Second, there has been a lack of investigation into the determinants of vitamin D concentration. For example, variables such as supplementary vitamin D intake may prevent serum levels from reflecting the patient’s exact physiological condition. Third, a detailed evaluation of plaque characteristics was not performed using appropriate methods, such as ultrasound or high-resolution magnetic resonance imaging. A larger prospective study with additional analyses that can compensate for the limitations mentioned above is warranted to understand the effect of vitamin D on prognosis after cervical CAS.

Low vitamin D levels were independently associated with successful carotid stent expansion. Our study suggests that the serum vitamin D level could be a biomarker for satisfactory carotid artery stenting.

Ethics Statement

The study protocol was approved by the Institutional Review Board (IRB) of Korea University Medical Center, Guro Hospital (IRB No. 2011GR0218). The requirement for informed consent was waived.

Availability of Data and Material

The data that support the findings of this study are available in the text.

Acknowledgments

None.

Sources of Funding

This research was supported by an R&D support grant (K2108031) from the Korea University Guro Hos-
pital, Republic of Korea.

Conflicts of Interest
No potential conflicts of interest relevant to this article was reported.

REFERENCES

1. Razzaque MS. The dualistic role of vitamin D in vascular calcifications. Kidney Int. 2011;79:708-714.
2. Kim DH, Meza CA, Clarke H, Kim JS, Hickner RC. Vitamin D and endothelial function. Nutrients. 2020;12:2575.
3. Kassi E, Adamopoulos C, Basdra EK, Papavassiliou AG. Role of vitamin D in atherosclerosis. Circulation. 2013;128:2517-2531.
4. Brott TG, Calvet D, Howard G, Gregson J, Algra A, Becquemin JP, et al. Long-term outcomes of stenting and endarterectomy for symptomatic carotid stenosis: a preplanned pooled analysis of individual patient data. Lancet Neurol. 2019;18:348-356.
5. Kang J, Hong JH, Kim BJ, Bae HJ, Kwon OK, Oh CW, et al. Residual stenosis after carotid artery stenting: effect on periprocedural and long-term outcomes. PLoS One. 2019;14:e0216592.
6. Seo WK, Lee JM, Park MH, Park KW, Lee DH. Cerebral microbleeds are independently associated with arterial stiffness in stroke patients. Cerebrovasc Dis. 2008;26:618-623.
7. Choi JY, Jung JM, Kwon DY, Park MH, Kim JH, Oh K, et al. Free fatty acid as an outcome predictor of atrial fibrillation-associated stroke. Ann Neurol. 2016;79:317-329.
8. Lee SH, Han JH, Jung I, Jung JM. Do thrombolysis outcomes differ between anterior circulation stroke and posterior circulation stroke? A systematic review and meta-analysis. Int J Stroke. 2020;15:849-857.
9. Sacco RL, Adams R, Albers G, Alberts MJ, Benavente O, Furie K, et al. Guidelines for prevention of stroke in patients with ischemic stroke or transient ischemic attack: a statement for healthcare professionals from the American Heart Association/American Stroke Association Council on Stroke: co-sponsored by the Council on Cardiovascular Radiology and Intervention: the American Academy of Neurology affirms the value of this guideline. Stroke. 2006;37:577-617.
10. Yadav JS, Wholey MH, Kuntz RE, Fayad P, Katzen BT, Mishkel GJ, et al. Protected carotid-artery stenting versus endarterectomy in high-risk patients. N Engl J Med. 2004;351:1493-1501.
11. Moore WS, Barnett HJ, Beebe HG, Bernstein EF, Brener BJ, Brott T, et al. Guidelines for carotid endarterectomy. A multidisciplinary consensus statement from the Ad Hoc Committee, American Heart Association. Circulation. 1995;91:566-579.
12. Giovannucci E, Liu Y, Hollis BW, Rimm EB. 25-hydroxyvitamin D and risk of myocardial infarction in men: a prospective study. Arch Intern Med. 2008;168:174-1180.
13. North American Symptomatic Carotid Endarterectomy Trial Collaborators, Barnett HJM, Taylor DW, Haynes RB, Sackett DL, Peerless SJ, et al. Beneficial effect of carotid endarterectomy in symptomatic patients with high-grade carotid stenosis. N Engl J Med. 1991;325:445-453.
14. Iakovou I, Mintz GS, Dangas G, Abizaid A, Mehran R, Kobayashi Y, et al. Increased CK-MB release is a "trade-off" for optimal stent implantation: an intravascular ultrasound study. J Am Coll Cardiol. 2003;42:1900-1905.
15. Arhuidese IJ, Rizwan M, Nejim B, Malas M. Outcomes of primary and secondary carotid artery stenting. Stroke. 2017;48:3086-3092.
16. Krupinski J, Catena E, Miguel M, Domenech P, Vila R, Mordon S, et al. D-dimer local expression is increased in symptomatic patients undergoing carotid endarterectomy. Int J Cardiol. 2007;116:174-179.
17. Schmidt R, Schmidt H, Pichler M, Enzinger C, Petrovic K, Niederkorn K, et al. C-reactive protein, carotid atherosclerosis, and cerebral small-vessel disease: results of the Austrian Stroke Prevention Study. Stroke. 2006;37:2910-2916.
18. Rubin MR, Rundek T, McMahon DJ, Lee HS, Sacco RL, Silverberg SJ. Carotid artery plaque thickness is associated with increased serum calcium levels: the Northern Manhattan study. Atherosclerosis. 2007;194:426-432.
19. Onufriak SJ, Bellasi A, Shaw LJ, Herzog CA, Cardarelli F, Wilson PW, et al. Phosphorus levels are associated with subclinical atherosclerosis in the general population. Atherosclerosis. 2008;195:242-243.
20. Suwelack B, Gerhardt U, Witta J, Hillebrandt U, Hohage H. Effect of parathyroid hormone levels on carotid intima-media thickness after renal transplantation. Am J Hypertens. 2001;14:1012-1018.
21. Cai J, Hatsuksami TS, Ferguson MS, Small R, Polissar NL, Yuan C. Classification of human carotid atherosclerotic lesions with in vivo multicontrast magnetic resonance imaging. Circulation. 2002;106:1368-1373.
22. Stary HC, Chandler AB, Dinsmore RE, Fuster V, Glagov S, Insull W Jr, et al. A definition of advanced types of atherosclerotic lesions and a histological classification of atherosclerosis. A report from the Committee on Vascular Lesions of the Council on Arteriosclerosis, American Heart Association. Circulation. 1995;92:1355-1374.

23. Sahara M, Kirigaya H, Oikawa Y, Yajima J, Nagashima K, Hara H, et al. Soft plaque detected on intravascular ultrasound is the strongest predictor of in-stent restenosis: an intravascular ultrasound study. Eur Heart J. 2004;25:2026-2033.

24. Fuster V, Badimon L, Badimon JJ, Chesebro JH. The pathogenesis of coronary artery disease and the acute coronary syndromes (2). N Engl J Med. 1992;326:310-318.

25. Saam T, Hetterich H, Hoffmann V, Yuan C, Dichgans M, Poppert H, et al. Meta-analysis and systematic review of the predictive value of carotid plaque hemorrhage on cerebrovascular events by magnetic resonance imaging. J Am Coll Cardiol. 2013;62:1081-1091.

26. McNally JS, Burton TM, Aldred BW, Kim SE, McLaughlin MS, Eisenmenger LB, et al. Vitamin D and vulnerable carotid plaque. AJNR Am J Neuroradiol. 2016;37:2092-2099.

27. Skultetyova D, Filipova S, Riecansky I, Skultety J. The role of angiotensin type 1 receptor in inflammation and endothelial dysfunction. Recent Pat Cardiovasc Drug Discov. 2007;2:23-27.

28. Virmani R, Farb A, Burke AP. Coronary angioplasty from the perspective of atherosclerotic plaque: morphologic predictors of immediate success and restenosis. Am Heart J. 1994;127:163-179.

29. Finet G, Weissman NJ, Mintz GS, Satler LF, Kent KM, Laird JR, et al. Mechanism of lumen enlargement with direct stenting versus predilatation stenting: influence of remodeling and plaque characteristics assessed by volumetric intracoronary ultrasound. Heart. 2003;89:84-90.

30. Wang J, Zhou JJ, Robertson GR, Lee VW. Vitamin D in vascular calcification: a double-edged sword? Nutrients. 2018;10:652.

31. Barrett HE, Cunnane EM, Hidayat H, O Brien JM, Kavanagh EG, Walsh MT. Calcification volume reduces stretch capability and predisposes plaque to rupture in an in vitro model of carotid artery stenting. Eur J Vasc Endovasc Surg. 2017;54:431-438.