Sonographic findings associated with stenosis progression and vascular complications in moyamoya disease

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OBJECTIVE The progression of arterial stenosis in patients with moyamoya disease (MMD) has variable courses and an unclear mechanism. The authors hypothesized that elevated wall shear stress (WSS) at the terminal internal carotid artery (ICA) and proximal middle cerebral artery (MCA) may facilitate MMD progression. They indirectly evaluated the relative magnitude of WSS (WSS value [WSSV]) with MR angiography (MRA) and transcranial Doppler to determine its predictive value for stenosis progression (SP) and the development of vascular complications.

METHODS Thirty-one medically treated patients (58 hemispheres and 95 nonoccluded vessels) were analyzed with serial MRA (median follow-up 23 months). The parameters studied were SP, SP rates (SPRs) for individual ICAs/MCAs, and their mean values from the ipsilateral hemispheres as mean SP (MSP) and MSP rates (MSPRs). Significant progression was defined as decrements of $\geq 20\%$ for SP and MSP and $\geq 10\%$ /year for SPR and MSPR. The development of vascular complications in relevant hemispheres was also recorded. The WSSV (dyne/cm²) was defined as the shear rate multiplied by blood viscosity.

RESULTS After adjusting the initial stenosis degree and MRA stage of MMD, an SP of $\geq 20\%$ and an SPR of $\geq 10\%$ /year were associated with the highest-quartile WSSVs for all individual vessels and for MCAs and ICAs separately. For each hemisphere, an MSP of $\geq 20\%$ and an MSPR of $\geq 10\%$ /year were associated with the highest-quartile mean WSSVs. Furthermore, significant SP was highly correlated with vascular complications, and the highest-quartile mean WSSV was independently associated with vascular complications in relevant hemispheres.

CONCLUSIONS An elevated WSSV is an independent predictor for SP and vascular complications in nonoccluded MMD.

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KEY WORDS moyamoya disease; transcranial Doppler; angiography; progression; vascular remodeling; wall shear stress; vascular disorders

MOYAMOYA disease (MMD) is an idiopathic, progressive arteriopathy of the terminal internal carotid artery (ICA) and proximal portions of its branches, including the middle cerebral artery (MCA) and the anterior cerebral artery.^{8,16,25} Stenosis progression (SP) in patients with MMD occurs frequently.¹⁷ Various factors

regarding vascular remodeling, including matrix metalloproteinases (MMPs) and their natural inhibitors (tissue inhibitors of metalloproteinases [TIMPs]), are known to be altered in MMD,^{2,5,8,13,14,16,19,25,30} implying that dysregulation of the vascular remodeling response to various stressors could be the mechanism of SP.^{8,16,25} Furthermore,

ABBREVIATIONS BV = blood viscosity; DR = diameter ratio; ICA = internal carotid artery; IQR = interquartile range; MCA = middle cerebral artery; MFV = mean flow velocity; MMD = moyamoya disease; MMP = matrix metalloproteinase; MRA = MR angiography; MSP = mean stenosis progression; MSPR = MSP rate; MWSSV = mean wall shear stress value; ROC = receiver operating characteristic; SD = stenosis degree; SP = stenosis progression; SPR = SP rate; SR = shear rate; TCD = transcranial Doppler; TIMP = tissue inhibitor of metalloproteinase; TOF = time-of-flight; WSS = wall shear stress; WSSV = WSS value.

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elevated endothelial wall shear stress (WSS), a force generated by the blood flow nearest to the vessel wall and imposed on the vascular endothelium, can significantly alter the levels and activities of these factors and consequently promote vascular remodeling.^{4,6,23,26} Considering that the WSS is highest at the primary pathological sites of MMD, the terminal ICA and its proximal branches, we hypothesized that elevated WSS may play an important pathophysiological role in the progression of MMD.^{14,29}

In terms of measuring the magnitude of WSS, numerous studies indirectly calculated the relative WSS value (WSSV) with sonography by measuring arterial diameter and centerline flow velocity.^{10,11} Transcranial Doppler (TCD) measures the velocity of blood flow in basal cerebral arteries and can estimate arterial stenosis.^{1,28,31} Therefore, TCD can be useful in diagnosing and staging MMD.¹⁸ TCD also precisely locates the site of arterial stenosis, so we can estimate the shear rate (SR) at a relevant site by using both TCD and MR angiography (MRA). In this study, we hypothesized that for nonoccluded vessels in patients with MMD, an elevated WSSV could be an independent predictor for SP. Therefore, we evaluated WSSVs with TCD and MRA to determine their relationship with SP and vascular complications.

Methods

Study Population and Follow-Up

We identified 106 patients older than 18 years who were diagnosed with MMD between 2003 and 2012 at Seoul National University Hospital. MMD was diagnosed by digital subtraction angiography according to current diagnostic criteria.¹⁵ To evaluate the factors associated with arterial stenosis, patients who met the following conditions were subjected to further analysis: 1) at least 1 vessel of the distal ICA and the proximal MCA of each hemisphere not occluded; 2) baseline MRI/MRA and TCD evaluations with an interval of less than 2 weeks; 3) favorable temporal windows for TCD and adequate TCD data; 4) follow-up MRI/MRA with an interval of more than 12 months; 5) no other potential causes of moyamoya syndrome; and 6) no surgical treatment for MMD until the follow-up MRI/MRA (because such a surgery might have significantly altered the hemodynamics of basal cerebral arteries). As a result, 19 patients with complete occlusion of both the MCA and the ICA, 26 patients with inadequate follow-up MRI/MRA, and 30 patients who had received surgical treatment were excluded. Thirty-one patients (58 hemispheres and 95 nonoccluded moyamoya vessels [57 ICAs and 38 MCAs]) were enrolled in our study. Clinical, laboratory, demographic, radiographic, and sonographic profiles of each patient were compiled. Vascular complications, such as cerebral hemorrhage and ischemia, that were confirmed by MRI were noted also. Approval to conduct this study was received from the Seoul National University Hospital Institutional Review Board. All medical records/information were anonymized and deidentified before analysis.

Analysis MRI and MRA

Initial and follow-up MRI/MRA was performed by us-

ing either a 1.5-T or a 3.0-T imaging system (Siemens Verio) according to an MRI protocol that included FLAIR, gradient echo, time-of-flight (TOF) angiography, and contrast-enhanced MRA with or without diffusion-weighted imaging. Stages of arterial stenosis in each hemisphere were scored from 0 to 10 by summing the scores of the ICA, MCA, anterior cerebral artery, and posterior cerebral artery according to a method introduced by Houkin et al.⁹ (0, normal; 1, stenosis of C_1/M_1 ; 2, focal occlusion of C_1/M_1 ; and 3, total nonvisualization of C_1/M_1 [for the ICA and MCA]; or 0, normal; 1, stenosis/focal occlusion of A_2/P_2 ; and 2, total nonvisualization of A_2/P_2 [for the anterior cerebral artery and posterior cerebral artery]). To evaluate the degree of stenosis precisely, we measured the arterial diameter of each nonoccluded vessel in the 4-times-magnified view of TOF angiography sequences at the most stenosed portion in the proximal M_1 segment for the MCA and the ophthalmic segment (C_6) for the ICA. Source images of TOF MRI sequences were also reviewed. Because vessel stenosis in patients with MMD is longitudinal, the diameter ratio (DR) was adopted for evaluating the relative degrees of stenosis. The DR was calculated by dividing the arterial diameter by the reference value, which is the proximal basilar artery diameter evaluated at the level just distal to the anterior inferior cerebellar artery branching.²⁴ The stenosis degree (SD) was calculated by subtracting the DR from 1 ($SD = 1 - DR$). The factors for disease progression were defined as follows: SP (percentage) as the increment of SD from the initial visit to the follow-up, and SP rate (SPR [percentage/year]) as SP divided by the time interval between the initial and follow-up images. The mean values of these factors from the ipsilateral hemispheres (ICAs/MCAs) were also calculated to evaluate the integrated SP of each hemisphere as follows: mean SP (MSP [percentage]) and MSP rate (MSPR [percentage/year]). If one of the vessels was occluded, values from the remaining non-occluded vessel were regarded as the mean values. Significant SP was defined for each parameter as follows: a progression of 20% or more for SP ($SP \geq 20\%$) and MSP ($MSP \geq 20\%$) and a progression of 10% or more per year for the SPR ($SPR \geq 10\%/year$) and MSPR ($MSPR \geq 10\%/year$).

Sonographic Analysis

Basal cerebral arteries were evaluated sonographically by a 2-MHz pulsed Doppler instrument according to standard protocol. The studies were conducted by a skilled sonographer and read by 2 stroke specialists who were blinded to the clinical data. TCD data, including mean flow velocity (MFV [cm/sec]) and pulsatility index, were obtained at the level of the distal portion of the ICA (from a transorbital window with an insonation depth of 65–80 mm, by identifying a receding flow proximal to the ICA bifurcation) and the proximal portion of the MCA (from a temporal window with an insonation depth of 65–70 mm, by identifying an approaching flow approximately 10–15 mm distal to the ICA bifurcation).²²

Evaluation of Endothelial Shear Stress

The WSSV (dyne/cm²) was calculated by multiplying the SR and blood viscosity (BV). Assuming that the termi-

nal ICA and proximal MCA are circular pipes and that the velocity profile of the flow is parabolic, SR (γ) could be defined as follows:

$$\gamma \text{ (sec}^{-1}\text{)} = 8 \times \text{MFV}/D,$$

where MFV and D are the mean flow velocity and lumen diameter, respectively.¹¹ For BV (μ), considering the shear thinning effect of blood flow, we adopted the Carreau-Yasuda model:

$$\mu \text{ (in cP)} = \mu_{\infty} + (\mu_0 - \mu_{\infty})[1 + (\lambda_{cy}\gamma)^a]^{(n-1)/a},$$

where μ is the viscosity for SR γ ; μ_{∞} is the viscosity for infinite SR; μ_0 is the viscosity at 0 SR; and λ_{cy} , a , and n are fitting parameters.³ For a valid blood-mimicking fluid, KSCN-X solution, these parameters have the following values: $\mu_0 = 22$ cP; $\mu_{\infty} = 2.2$ cP; $a = 0.644$; $n = 0.392$; and $\lambda_{cy} = 0.110$ sec. Given that the BV increases exponentially with increasing hematocrit levels, correction for the variations in BV according to the hematocrit level is necessary. We used the method proposed by Box et al.³ to replace the parameter n with the hematocrit level. Mean WSSVs from the ipsilateral ICAs/MCAs were also calculated and defined as mean WSSV (MWSSV) (dyne/cm²). The Reynolds number was calculated according to the following equation:

$$R = (\rho \times \text{MFV} \times r)/\mu,$$

where ρ is the blood density (assumed to be 1.060×10^3 kg/m³), MFV is the mean velocity of the blood (m/sec), r is the radius of the vessel (mm), and μ is the BV (cP). A Reynolds number of < 1000 is usually considered characteristic of laminar flow.⁷ The luminal irregularity, or curvature of the vessels, was not taken into account in the evaluation of the WSSV.

Statistical Analysis

IBM SPSS Statistics version 21.0 for Windows was used for statistical analysis. Student t-tests were conducted for interval variables, and chi-square tests were used for dichotomous or qualitative variables. Variables with a p value < 0.10 were included in the multivariate analysis. For multivariate analysis, a logistic regression test was used. A receiver operating characteristic (ROC) curve was used to evaluate the efficacy of the WSSV to predict SP. Statistical significance was set at $p < 0.05$.

Results

Characteristics of the Study Population

The median follow-up duration was 23 months (range 12–60 months, interquartile range [IQR] 15–35 months). Eight patients (25.8%) were male, and the mean age was 46.0 ± 11.5 years. Clinical and demographic profiles are listed in Table 1. For the 58 hemispheres, the initial mean SD ($\pm$ standard deviation) was $27.1\% \pm 17.8\%$. The median MRA stages were 3 (IQR 1–4) for each hemisphere and 5 (IQR 4–8) for the sum of bilateral hemispheres. The median follow-up MRA stage was 4 (IQR 2–5), and progression of stage was noted in 9 patients (15.5%) for each hemisphere. Ten vascular complications (17.2%; 7 ische-

TABLE 1. Demographic and laboratory data (n = 31)*

Demographic or Lab Info	Value
Age in yrs	46.0 ± 11.5
Male sex	8 (25.8)
Hypertension	10 (32.3)
Diabetes	2 (6.5)
Hyperlipidemia	4 (12.9)
Smoking	1 (3.2)
HbA1c (%)	5.7 ± 0.5
Total cholesterol (mg/dl)	170.0 ± 28.4
LDL cholesterol (mg/dl)	98.7 ± 28.9
Hematocrit (%)	39.9 ± 4.1

HbA1c = glycated hemoglobin; Info = information; LDL = low density lipoprotein.

* Data are reported as mean $\pm$ standard deviation or number (%).

mia and 3 hemorrhage) developed during follow-up with a mean delay of 21.9 ± 9.7 years (Table 2).

Stenosis Progression

For each vessel, the initial mean SD was 38.0 ± 17.8 (20.2 ± 18.7 for ICAs, 42.7 ± 21.9 for MCAs). The SP and SPR were $5.9\% \pm 21.8\%$ ($0.01\% \pm 19.09\%$ for ICAs, $14.65\% \pm 23.08\%$ for MCAs) and $3.5\% \pm 17.2\%$ /year ($0.21\% \pm 15.29\%$ /year for ICAs, $8.52\% \pm 19.01\%$ /year for MCAs), respectively. Significant SP for each parameter was reported as follows: 22 (23.2%) for an SP $\geq 20\%$ (7 [12.3%] for ICAs, 15 [39.5%] for MCAs) and 26 (27.4%) for an SPR $\geq 10\%$ /year (10 [17.5%] for ICAs, 16 [42.1%] for MCAs). For each hemisphere, the MSP and MSPR were $5.8\% \pm 16.1\%$ and $3.4\% \pm 12.4\%$ /year, respectively. Significant SP for each parameter was as follows: 10 (17.2%) for an MSP $\geq 20\%$ and 12 (20.7%) for an MSPR $\geq 10\%$ /year. MCAs showed more severe initial SD and more frequent significant SP than ICAs (Tables 2 and 3).

TABLE 2. Radiological profiles for hemispheres*

Variable	Value
MWSSV (dyne/cm ²)	13.5 ± 7.3
Initial mean SD (%)	27.1 ± 17.8
MRA stage	3 (1–4)
MRA stage >3	36 (62.0)
Follow-up mean SD (%)	32.6 ± 19.4
Follow-up MRA stage	4 (2–5)
MRA stage progression	9 (15.5)
MSP (%)	5.8 ± 16.1
MSP $\geq 20\%$	10 (17.2)
MSPR (%/yr)	3.4 ± 12.4
MSPR $\geq 10\%$ /yr	12 (20.7)
Hemorrhage	3 (5.2)
Ischemia	7 (12.1)

* Data are reported as mean $\pm$ standard deviation, median (IQR), or number (%) of hemispheres (n = 58).

Profiles of WSS

In the sonographic analyses, the MWSSVs were 12.3 ± 8.5 dyne/cm² for ICAs and 15.4 ± 9.7 dyne/cm² for MCAs, and the MWSSV for each hemisphere was 13.5 ± 7.3 dyne/cm² (Tables 2 and 3). The Reynolds number ranged from 35.0 to 683.0, indicating that blood flows could be regarded as laminar. To evaluate the effect of an elevated WSSV in the progression of vessel stenosis, we selected vessels/hemispheres of which the WSSV/MWSSV were in the highest-quartile group (highest-quartile WSSV/MWSSV). To adjust the effect of initial SD with the WSSV or the progression of vessel stenosis, we evaluated the relations of progression factors with factors such as having an initial SD value higher than the median (upper-half SD) or higher than the third quartile (highest-quartile SD) and advanced hemispheric MRA stage (MRA stage ≥ 4), and we adjusted them for later analyses.

Factors Associated With SP in Individual Nonoccluded Vessels

In the univariate analysis for factors related to significant SP in MCAs, the highest-quartile WSSV was significantly associated with an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year, whereas the highest-quartile SD was associated with an SPR $\geq 10\%$ /year. No conventional cerebrovascular risk factor was associated with SP. After adjusting for factors associated with the initial SD, multivariate analysis showed that a WSSV in the highest quartile was the only prognostic factor for an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year. In the univariate analysis of ICAs, a WSSV in the highest quartile was associated significantly with an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year, whereas factors regarding the initial SD or cardiovascular risk factors were insignificant. In the multivariate analysis, a WSSV in the highest quartile was confirmed to be the predictive factor for an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year. In the univariate analysis of all individual vessels (MCAs and ICAs combined), a WSSV

in the highest quartile was significantly associated with an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year, whereas an SD in the highest quartile was associated with an SP $\geq 20\%$. Moreover, MCAs, more so than ICAs, were associated with an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year. Multivariate analysis showed that a WSSV in the highest quartile was the independent prognostic factor for an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year. MCAs, more so than ICAs, remained significantly associated with an SP $\geq 20\%$ and an SPR $\geq 10\%$ /year (Table 4).

Factors Associated With SP in Individual Hemispheres

In the univariate analysis for factors related to significant SP in each hemisphere, an MWSSV in the highest quartile was significantly associated with an MSP $\geq 20\%$ and an MSPR $\geq 10\%$ /year, whereas hyperlipidemia was also associated with an MSP $\geq 20\%$. No other conventional cerebrovascular risk factors or initial SD factors were significantly associated with SP. Multivariate analysis revealed that an MWSSV in the highest quartile was the only prognostic factor for an MSP $\geq 20\%$ and an MSPR $\geq 10\%$ /year (Table 5). The durations of follow-up were comparable between the group with an MSP $\geq 20\%$ and the group with an MSP $< 20\%$, and the group with an MSPR $< 10\%$ /year had a longer follow-up duration than the group with an MSPR $\geq 10\%$ /year. Furthermore, groups with significant hemispheric SP (MSP $\geq 20\%$ and MSPR $\geq 10\%$ /year) showed more frequent development of vascular complications than the groups without it. The times until complication development were comparable between the groups (Table 6).

Factors Associated With Vascular Complications in Individual Hemispheres

In the univariate analysis for factors related to the development of vascular complications in relevant hemispheres, an MWSSV in the highest quartile was significantly associated with the development of vascular complications. No other conventional cerebrovascular risk factors or initial SD factors had a significant association. Multivariate analysis revealed that an MWSSV in the highest quartile was the only predictor for vascular complications (Table 7).

ROC Curve Analysis

In ROC curve analysis for MCAs, a WSSV in the highest quartile predicted an SP $\geq 20\%$ with a sensitivity of 73.3% and a specificity of 69.6%, and an SPR $\geq 10\%$ /year with a sensitivity of 81.3% and a specificity of 68.2%. For ICAs, a WSSV in the highest quartile predicted an SP $\geq 20\%$ with a sensitivity of 71.4% and a specificity of 64.0%, and an SPR $\geq 10\%$ /year with a sensitivity of 80.0% and a specificity of 64.0%. For all individual vessels combined, a WSSV in the highest quartile predicted an SP $\geq 20\%$ with a sensitivity of 81.8% and a specificity of 71.2%, and an SPR $\geq 10\%$ /year with a sensitivity of 80.8% and a specificity of 71.0%. For each hemisphere, an MWSSV in the highest quartile predicted an MSP $\geq 20\%$ with a sensitivity of 76.5% and a specificity of 69.2%, and an MSPR $\geq 10\%$ /year with a sensitivity of 63.6% and a specificity of 58.7% (Fig. 1).

TABLE 3. WSS and SP profiles for vessels*

Variable	ICA (n = 57)	MCA (n = 38)	p Value
Diameter (mm)	2.3 $\pm$ 0.6	1.7 $\pm$ 0.6	<0.001†
MFV (cm/sec)	70.4 $\pm$ 45.3	123.3 $\pm$ 65.8	<0.001†
PI	0.7 $\pm$ 0.2	0.6 $\pm$ 0.1	0.002†
SR (sec ⁻¹)	285.7 $\pm$ 299.8	377.7 $\pm$ 307.7	0.153
WSSV (dyne/cm ²)	12.3 $\pm$ 8.5	15.4 $\pm$ 9.7	0.109
Reynolds number	180.9 $\pm$ 132.2 (41.2–683.0)	223.3 $\pm$ 127.0 (35.0–493.2)	0.084
Initial SD (%)	20.2 $\pm$ 18.7	42.7 $\pm$ 21.9	0.002†
Follow-up SD (%)	20.2 $\pm$ 25.1	57.3 $\pm$ 35.0	
SP (%)	0.0 $\pm$ 19.1	14.7 $\pm$ 23.1	0.002†
SP $\geq 20\%$	7 (12.3)	15 (39.5)	0.002†
SPR (%/yr)	0.2 $\pm$ 15.3	8.5 $\pm$ 19.0	0.027†
SPR $\geq 10\%$ /yr	10 (17.5)	16 (42.1)	0.008†

PI = pulsatility index.

* Data are reported as mean $\pm$ standard deviation (range) or number (%) of vessels (n = 95).

† Statistically significant (p < 0.05).

TABLE 4. Factors associated with SP for nonoccluded vessels

Vessel & Factor	SPR ≥20%				SPR ≥10%/yr			
	Univariate Analysis		Multivariate Analysis		Univariate Analysis		Multivariate Analysis	
	p Value	OR (95% CI)	p Value	OR (95% CI)	p Value	OR (95% CI)	p Value	OR (95% CI)
MCA (n = 38)								
Male sex	0.722	1.417 (0.342–5.866)			0.471	2.040 (0.493–8.446)		
Hypertension	1.000	1.030 (0.236–4.504)			0.713	1.545 (0.361–6.610)		
Diabetes	0.509	NA			1.000	1.400 (0.080–24.201)		
Hyperlipidemia	0.490	NA			0.171	NA		
Smoker	1.000	NA			1.000	NA		
Initial SD (upper half)	0.097	3.111 (0.797–12.140)	0.336		0.189	2.407 (0.642–9.027)	0.708	
Initial SD (highest quartile)	0.115	4.444 (0.903–21.874)	0.331		0.021*	7.778 (1.342–45.090)	0.128	
MRA stage >3	0.145	0.239 (0.043–1.321)			1.000	1.212 (0.295–4.982)		
Highest-quartile WSSV	0.016*	9.188 (1.565–53.934)	0.014*	9.187 (1.565–53.934)	0.002*	21.000 (2.252–195.816)	0.008*	21.000 (2.252–195.816)
ICA (n = 57)								
Male sex	1.000	1.138 (0.196–6.600)			0.713	1.250 (0.278–5.619)		
Hypertension	0.667	1.750 (0.348–8.795)			0.709	1.571 (0.383–6.444)		
Diabetes	1.000	NA			0.446	2.500 (0.204–30.605)		
Hyperlipidemia	0.252	2.933 (0.462–18.626)			0.137	3.600 (0.698–18.555)		
Smoker	1.000	NA			1.000	NA		
Initial SD (upper half)	1.000	0.750 (0.152–3.701)	0.215		0.504	1.705 (0.425–6.836)	0.772	
Initial SD (highest quartile)	0.346	2.659 (0.516–13.704)	0.492		0.694	1.403 (0.309–6.360)	0.243	
MRA stage >3	1.000	0.955 (0.193–4.717)			0.735	1.350 (0.344–5.301)		
Highest-quartile WSSV	0.033	5.333 (1.025–27.758)	0.047*	5.333 (1.025–27.758)	0.010*	7.313 (1.671–32.003)	0.008*	7.312 (1.671–32.003)
All individual vessels (n = 95)								
Male sex	0.593	1.326 (0.470–3.746)			0.331	0.619 (0.610–4.298)		
Hypertension	0.783	1.156 (0.412–3.238)			0.500	1.393 (0.531–3.658)		
Diabetes	0.587	NA			0.612	1.833 (0.289–11.650)		
Hyperlipidemia	0.233	2.481 (0.632–9.746)			0.130	3.048 (0.803–11.569)		
Smoker	1.000	NA			0.559	NA		
Initial SD (upper half)	0.303	1.657 (0.630–4.355)	0.984		0.149	1.961 (0.780–4.929)	0.420	
Initial SD (highest quartile)	0.037*	2.918 (1.041–8.176)	0.177		0.061	2.692 (0.996–7.276)	0.072	
MRA stage >3	0.094	0.398 (0.144–1.197)	0.101		0.944	1.034 (0.409–2.615)		
MCA vs ICA	0.001*	5.083 (1.797–14.381)	0.002*	6.203 (1.943–19.804)	0.009*	3.418 (1.337–8.736)	0.005*	4.996 (1.617–15.435)
Highest-quartile WSSV	0.004*	4.214 (1.521–11.673)	0.001*	6.930 (2.105–22.820)	<0.001*	7.778 (2.744–22.045)	<0.001*	10.616 (3.248–34.700)

NA = not available.

* Statistically significant ($p < 0.05$).

TABLE 5. Factors associated with SP in 58 hemispheres*

Factor	MSP $\geq 20\%$			MSPR $\geq 10\%/yr$		
	Univariate Analysis		Multivariate Analysis	Univariate Analysis		Multivariate Analysis
	p Value	OR (95% CI)	p Value	OR (95% CI)	p Value	OR (95% CI)
Male sex	0.713	1.250 (0.278–5.619)		1.600 (0.442–5.796)	0.507	
Hypertension	0.478	0.484 (0.092–2.555)		1.115 (0.317–3.921)	1.000	
Diabetes	1.000	NA		1.429 (0.120–16.998)	1.000	
Hyperlipidemia	0.025	7.167 (1.407–36.504)	0.147	1.850 (0.384–8.914)	0.422	
Smoker	1.000	NA		NA	1.000	
Initial mean SD (upper half)	0.504	1.705 (0.425–6.836)	0.995	1.816 (0.548–6.019)	0.326	0.284
Initial mean SD (highest quartile)	0.694	1.403 (0.309–6.360)	0.973	2.833 (0.781–10.227)	0.161	0.311
MRA stage >3	0.160	0.261 (0.050–1.361)	0.082	0.550 (0.160–1.886)	0.339	0.805
Highest-quartile MWSSV	0.002*	11.667 (2.474–55.020)	0.002*	11.667 (2.474–55.020)	0.045*	7.877 (1.392–44.565)

* Statistically significant ($p < 0.05$).

Discussion

In patients with MMD, before vessels became occluded, a WSSV in the highest quartile was associated with the absolute magnitude (SP) and rate (SPR) of SP in each vessel and also with the absolute magnitude (MSP) and rate (MSPR) of SP in each hemisphere. Furthermore, significant SP was highly associated with the development of symptoms, and a hemispheric MWSSV in the highest quartile was an independent predictor of vascular complications in the relevant hemispheres. This relationship was independent of initial SD, MR stage of MMD, duration of follow-up, and other vascular risk factors. This result might be explained by an altered vascular remodeling response to elevated WSS; thus, the WSSV according to TCD might be an important indicator of MMD progression.

When the vessel is not stenosed and blood flow is undisturbed, WSS normally keeps endothelial cells in a stable state by downregulating inflammatory cytokines (e.g., interleukin 1, interleukin 4, and tumor necrosis factor- α) and growth factors (e.g., platelet-derived growth factor, fibroblast growth factor-2, and insulin-like growth factor-1).^{4,8,16,23} This noninflammatory/nonproliferative state results in the suppression of basement membrane-degrading MMPs (MMP-2 and MMP-9) and inhibition of smooth-muscle cell migration into the intima.^{6,8,12,16,20,21,23} However, in a stenosed vessel, a decrease in the diameter likely increases the WSS. Elevated WSS causes endothelial cells and smooth-muscle cells to significantly overexpress MMP-2 and MMP-9, consequently inducing a disproportional increase of MMPs over TIMPs and thereby degrading the extracellular matrix and promoting the migration and proliferation of vascular smooth-muscle cells, which results in adaptive vascular remodeling.²⁶

In patients with MMD, SP is accompanied by fibrocellular thickening of the intima with an increased number of smooth-muscle cells, undulation of the elastic lamina, and attenuation of the media,^{16,25} which implies a possibility that the altered behaviors of MMPs and TIMPs play a significant pathophysiological role in the SP of MMD. There have been numerous studies regarding the altered expression of MMPs and TIMPs in patients with MMD. Overexpression of MMP-9^{2,5,13} and reduced expression of MMP-3, TIMP-1, and TIMP-2^{2,13,30} have been documented in patients with MMD. Genetic studies of familial MMD have found polymorphisms in the MMP-3 gene¹⁹ and the TIMP-2 promoter gene.¹⁴ These alterations in MMP expression/function might result in immoderate extracellular matrix degradation/deposition and smooth-muscle cell proliferation/migration in response to elevated WSS. The excessive thickening of the intima and progressive vascular narrowing in patients with MMD are possible results of this dysregulation in the vascular remodeling response to high WSS in the terminal ICA and its proximal branches, where the flow is considerably fast and branches are in a very acute angle (proximal MCA). Considering that the initial SD was not significantly associated with SP in our study, WSS elevation might be an independent driving force behind the progression of MMD and not simply a consequence of vessel stenosis.

Our finding that MCAs were more associated with SP than ICAs is also interesting. Considering the relatively

TABLE 6. Comparison of follow-up duration, vascular complication frequency, and time to complication development in groups with and those without significant SP*

Factor	Hemispheres w/ MSP $\geq 20\%$	Hemispheres w/ MSP $< 20\%$	p Value	Hemispheres w/ MSPR $\geq 10\%/yr$	Hemispheres w/ MSPR $< 10\%/yr$	p Value
Follow-up duration (mos)	22.4 $\pm$ 10.6	27.3 $\pm$ 14.0	0.231	19.8 $\pm$ 9.3	28.2 $\pm$ 14.0	0.021†
Vascular complication (%)	80.0	4.2	< 0.001 †	50.0	8.7	< 0.001 †
Time to complication development (mos)	22.1 $\pm$ 10.7	21.0 $\pm$ 7.1	0.893	18.8 $\pm$ 10.3	26.5 $\pm$ 7.7	0.217

* Data are reported as mean $\pm$ standard deviation or percentage.† Statistically significant ($p < 0.05$).**TABLE 7. Factors associated with vascular complication for 58 hemispheres**

Factor	Vascular Complication			
	Univariate Analysis		Multivariate Analysis	
	p Value	OR (95% CI)	p Value	OR (95% CI)
Male sex	0.439	2.000 (0.482–8.306)		
Hypertension	1.000	0.857 (0.195–3.764)		
Diabetes	1.000	NA		
Hyperlipidemia	0.702	0.651 (0.071–5.968)		
Smoker	1.000	NA		
Initial mean SD (upper half)	0.164	2.758 (0.636–11.956)	0.082	
Initial mean SD (highest quartile)	0.233	2.533 (0.598–10.738)	0.287	
MRA stage > 3	0.160	0.250 (0.048–1.301)	0.069	
Highest-quartile MWSSV	0.002*	11.667 (2.474–55.020)	0.002*	15.881 (2.719–82.770)

* Statistically significant ($p < 0.05$). **FIG. 1.** Predictive power of a high WSS for SP:ROC curves. ROC curves of WSSVs for parameters of significant SP ($SP \geq 20$ and $SPR \geq 10$ for ICAs/MCAs/all vessels, and $MSP \geq 20$ and $MSPR \geq 10$ for hemispheric means) are shown. Figure is available in color online only.

early disease stages of the study population, we can infer that the proximal MCA might usually be involved before the distal ICA in the early stage of MMD. Additional studies aimed at replicating our findings and determining the underlying mechanism should be conducted.

There are several limitations of our study. First, we indirectly estimated that the SR of blood flow would be Newtonian and did not take vessel curvature into account. Therefore, we adopted an indirect term, WSSV, to indicate the relative magnitude of the actual WSS. Computational flow dynamic studies using 3D angiography are necessary to evaluate the actual WSS at the site of interest. This direct estimation is especially important in investigating WSS profiles of turbulent flow characteristics, such as those of intracranial aneurysms.²⁷ However, in clinical contexts in which laminar flow could be assumed, numerous studies sonographically measured the relative magnitude of WSS and performed quantitative analysis to establish its pathophysiological role in the development of various vascular diseases.^{10,11} The parabolic velocity profile was assumed in those studies, and velocity at the centerline of flow was evaluated. Considering that the ophthalmic segment of the ICA and the proximal M₁ of the MCA are relatively large and straight, that the flow characteristics in this study were laminar with uniformly low Reynolds numbers, and that the MFV can be estimated precisely with TCD,¹ the relative magnitude of WSS evaluated in this study might have sufficient validity. Second, we used TOF sequences to estimate SDs, which might have overestimated the SDs in cases of highly stenosed vessels. However, the highest initial SD was 72.4% (the highest follow-up SD was 83.4%), and only 2 vessels had an SD > 70% (6 vessels had a follow-up SD ≥ 70%), which indicates that the initial and follow-up SDs might have been evaluated appropriately in most cases. Third, the number of patients in this study was small, which might weaken the statistical certainty. This small study population might be related to the rarity of MMD and the high frequency of surgical intervention. Fourth, TCD is an evaluation tool that depends highly on the technician. In addition, our retrospective design and relatively short follow-up duration (median 23 months) might also be limitations of our study. Prospective studies aimed at confirming and extending the retrospective findings of different Doppler laboratories might be warranted.

Conclusions

In patients with MMD, elevated WSS is independently associated with SP and the development of vascular complications. Evidence from large prospective studies or from direct measurement of WSS is needed before applying these values in clinical practice.

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Disclosures

The authors report no conflict of interest concerning the materials or methods used in this study or the findings specified in this paper.

Author Contributions

Conception and design: Jung, KJ Lee, ST Lee, Roh. Acquisition of data: WJ Lee. Analysis and interpretation of data: WJ Lee, Kim, Chu. Drafting the article: WJ Lee. Critically revising the article: SK Lee, Roh. Reviewed submitted version of manuscript: all authors. Approved the final version of the manuscript on behalf of all authors: Jung. Statistical analysis: WJ Lee. Administrative/technical/material support: Kim, ST Lee. Study supervision: Jung, Chu, SK Lee.

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