Adenosine Diphosphate–Induced Platelet Aggregation Might Contribute to Poor Outcomes in Atrial Fibrillation–Related Ischemic Stroke

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Systemic atherosclerosis is involved in ischemic damages and cardioembolism after atrial fibrillation (AF)–related ischemic stroke (IS). Platelet activation is a critical factor in systemic atherosclerosis; however, there is little information regarding the role of platelet activation on the outcome of AF-related IS. We investigated the relationship between adenosine diphosphate (ADP)–induced platelet aggregation and the long-term outcomes of AF-related IS. We studied 249 patients who were exclusively treated with anticoagulation therapy after they had experienced AF-related IS. We evaluated their platelet function 5 days after admission to the hospital by using an optic platelet aggregometer test. We also assessed the prognoses of patients 90 days after the AF-related IS. Our results showed that ADP-induced platelet aggregation was positively correlated with CHA2DS2-VASc scores ($r = .285$, $P < .01$). Totally, 107 (43.0%) patients had a poor outcome at 90 days after IS. Univariate analysis showed that the following factors significantly contribute to a poor outcome: older age (odds ratio [OR] = 1.07, confidence interval [CI] 1.04-1.10, $P < .01$), a history of stroke (OR = 3.24, CI 1.61-6.53, $P < .01$), high scores on the National Institutes of Health Stroke Scale (NIHSS; OR = 1.25, CI 1.18-1.32, $P < .01$), increased white blood cell counts (OR = 1.12, CI 1.02-1.24, $P < .01$), high CHA2DS2-VASc scores (≥5, OR = 7.31, CI 3.36-15.93, $P = .025$), and the highest tertile of ADP-induced platelet aggregation (≥72%, OR = 3.17, CI 1.67-5.99, $P < .01$). Of these factors, high NIHSS scores (OR = 1.27, CI 1.20-1.36, $P < .01$), high CHA2DS2-VASc scores (OR = 4.69, CI 1.21-18.14, $P = .03$), and the highest tertile of ADP-induced platelet aggregation (OR = 2.49, CI 1.17-5.27, $P = .02$) were independently associated with a poor outcome at 90 days after IS. Therefore, our results suggest that platelet activation might affect the outcome of AF-related IS. **Key Words:** Platelet aggregation—atrial fibrillation—ischemic stroke—adenosine diphosphate.

Introduction

Atrial fibrillation (AF) is the most common cause of cardiac embolism, accounting for approximately 77% of the high-risk cardiac sources of embolism in ischemic stroke (IS).[1] The incidence of AF-related IS has increased considerably in recent years because of increases in the elderly population.[2] The risk of recurrence and mortality is higher after AF-related IS than other types of IS. CHADS2 is a scoring system that is used to select patients eligible for anticoagulation therapy, and it has a significant predictive value for mortality in patients with AF-related IS.[3,4] Recently, the CHA2DS2-VASc score was introduced in clinical practice. This new scoring system added the following predictors to the CHADS2 score: (1) vascular disease (myocardial infarction, complex aortic plaque, and peripheral artery disease), (2) age between 65 and 74 years, and (3) female sex. The prediction of thromboembolism with the

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CHA2DS2-VASc scoring system was modestly better than that with the CHADS2 score.[5]

Several studies have, thus far, reported that an increase in CHADS2/CHA2DS2-VASc scores was related to high mortality and poor outcomes in patients who experienced AF-related IS.[6-8] The exact mechanism responsible for the higher mortality rates associated with AF during stroke has not yet been elucidated. However, it can be presumed that these scoring systems indicate the likelihood of an AF-related IS and the burden of systemic atherosclerosis in AF.[9] Therefore, the presence of systemic atherosclerosis might increase the mortality rate after AF-related IS. Thus far, there has been little evidence to clarify the role of systemic atherosclerosis in AF-related IS.

Atherosclerosis is a systemic process, mediated by several factors, including endothelial cells, inflammatory cells, tissue factors, and platelets. Platelet activation is involved in all the key steps that comprise the progression of atherosclerosis.[10] Many studies have shown that platelet activation is a critical factor that determines the outcome of atherothrombosis, including myocardial infarction and stroke.[11,12] Therefore, we hypothesized that increased platelet activation could be related to the atheroma burden and potentiated ischemic damage in AF-related IS. To test this hypothesis, we investigated whether CHA2DS2-VASc scores correlate with the extent of ADP-induced platelet aggregation, and we then assessed whether ADP-induced platelet aggregation influenced the outcome in AF-related IS.

Subjects and Methods

The present study retrospectively recruited 3758 patients who were registered on the stroke registry of Dong-A University, Busan, Korea, between March 2000 and September 2012 after an acute ischemic stroke (AIS; within 72 hours of their ischemic events). We included the following patients in our study population: (1) those who had an AF-related stroke mechanism, (2) those who were not given any antiplatelet agents during the admission period, (3) those who could be followed-up 90 days after AIS, (4) and those in whom platelet function was evaluated according to the predefined method. Stroke neurologists assessed baseline National Institutes of Health Stroke Scale (NIHSS) for all the patients included in our study. On the day of admission, the medical history of each patient was recorded, and complete physical and neurologic examinations were performed. All patients were subject to a standard investigative protocol, including routine blood tests and transcranial Doppler ultrasonography, electrocardiography, and transthoracic echocardiography in selected cases. We calculated the pre-admission CHA2DS2-VASc score for each patient according to previously published data,[6-8] and patients were categorized in 3 groups based on their CHA2DS2-VASc score: 0-2, 3-4, and 5 or more.

For brain imaging studies, we examined the brain computed tomography and/or magnetic resonance imaging and magnetic resonance angiogram of all patients. The modified Rankin Scale was calculated at baseline and at 90 days after IS. A poor outcome was defined as a modified Rankin Scale score greater than 2 at 90 days after IS. This study was approved by the local ethics committee.

Optical Platelet Aggregometer

We evaluated the extent of platelet aggregation in our patients on day 5 after their admission to the stroke center. For this, 30 mL of whole blood was anticoagulated with 3.2% of sodium citrate. Platelet-rich plasma (PRP) was prepared by centrifugation at 160*g* for 10 minutes at room temperature. The platelet count of the PRP was adjusted to 200,000/mm^3 using platelet poor plasma obtained by centrifugation at 4000*g* for 5 minutes. For this, we used an optical aggregometer (Chrono Log, 560VS). Platelets were stimulated with arachidonic acid (.5 mg/mL) or adenosine diphosphate (ADP, 10 μM). We measured the extent of platelet aggregation for a period of 6 minutes after stimulation with either one of the agonists, AA or ADP. The results are expressed as the percent aggregation (percent aggregation is the estimated percentage difference in the light that is transmitted by PPP and PRP). This method has been verified previously.[13,14]

Statistical Analysis

Categorical variables were summarized according to counts and relative frequencies and numeric variables according to mean ± standard deviation or median (Q1-Q3). Patients were divided into 3 subgroups according to CHA2DS2-VASc scores (0-2, 3-4, ≥5). We used Pearson correlation coefficients to test for correlations between CHA2DS2-VASc scores and the extent of ADP-induced platelet aggregation. Clinical and laboratory findings were compared between the 3 groups. The significance of intergroup differences was assessed by chi-square analysis for categorical variables and analysis of variance or Kruskal–Wallis test for continuous variables. Post hoc analyses of differences between the 3 groups were performed with Scheffe and Bonferroni tests. Comparisons between good and poor outcomes at 90 days after IS were made using *t* tests for continuous variables and chi-square or Wilcoxon rank sum tests for categorical variables. The odds ratio (OR) for comparison of the 2 groups was summarized, together with the 95% confidence interval and *P* value, using logistic regression. The data for ADP-induced platelet aggregation were stratified based on tertiles. In addition, a multivariate model was created using a backward elimination method. The probability was set at .10 for removal. ORs were also adjusted for factors affecting the response variables. *P* values less than .05