Alkaline Phosphatase and Risk of Stroke Among Japanese: The Circulatory Risk in Communities Study (CIRCS)

Yuji Shimizu, MD, PhD,*† Hironori Imano, MD, PhD,* Tetsuya Ohira, MD, PhD,*† Akihiko Kitamura, MD, PhD,† Masahiko Kiyama, MD, PhD,† Takeo Okada, MD, PhD,† Yoshinori Ishikawa, MD,† Takashi Shimamoto, MD, PhD,† Kazumasa Yamagishi, MD, PhD,‡ Takeshi Tanigawa, MD, PhD,§ and Hiroyasu Iso, MD, PhD,* for the CIRCS Investigators

Although serum alkaline phosphatase (ALP) levels have been associated with mortality from all-cause and from either ischemic or hemorrhagic stroke, no study has been published of the associations between ALP and the incidence of stroke. We therefore examined the associations of ALP with risk of stroke among Japanese, stratified by drinking status because ALP is known as an enzyme affected by alcohol consumption. We conducted a prospective cohort study of 10,754 Japanese subjects (4098 men and 6656 women) aged 40-69 years and living in 4 communities under systematic surveillance for stroke incidence. During the 16-year follow-up, we documented 264 strokes (164 ischemic strokes and 69 hemorrhagic strokes) for men and 225 strokes (118 ischemic strokes and 89 hemorrhagic strokes) for women. There was a U-shaped association between ALP level and stroke incidence in both men and women, which was confined primarily to nondrinkers. For nondrinkers, higher ALP levels were associated with an elevated risk of ischemic stroke for men and of hemorrhagic stroke for women, whereas lower ALP levels were associated with elevated risks of ischemic and hemorrhagic strokes in both men and women. Our data indicate that not only higher, but also lower, serum ALP level may be a predictor for the risk of stroke in nondrinking men and women. **Key Words:** Alkaline phosphatase—CIRS—risk—incidence—Japanese.

Alkaline phosphatase (ALP) is an enzyme that catalyzes the hydrolysis of inorganic pyrophosphate,[1] an inhibitor of vascular calcification.[2] ALP is expressed in a variety of tissues, but its concentrations are notably high in bone, liver, and kidneys.[1] Previous prospective studies have identified an association between higher ALP levels and mortality from all causes and with cardiovascular disease in subjects with previous myocardial infarction[3] and previous stroke,[4] clinic populations,[5] and general populations.[3] Serum ALP levels are influenced by alcohol consumption,[6,7] and alcohol consumption has been positively associated with stroke incidence.[8]

From the *Public Health, Department of Social and Environmental Medicine, Graduate School of Medicine, Osaka University, Osaka, Japan; †Osaka Medical Center for Health Science and Promotion, Osaka, Japan; ‡Department of Public Health Medicine, Graduate School of Comprehensive Human Sciences and Institute of Community Medicine, University of Tsukuba, Tsukuba, Japan; and §Department of Public Health, Social Medicine and Medical Informatics, Ehime University Graduate School of Medicine, Toon, Japan.

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Address correspondence to Hiroyasu Iso, MD, PhD, Public Health, Department of Social and Environmental Medicine, Osaka University Graduate School of Medicine, Suita-shi, Osaka 565-0871, Japan. E-mail: iso@pbhel.med.osaka-u.ac.jp.

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However, no study of the associations between ALP and health outcome for subjects stratified by drinking status has been published to date. One prospective study reported an association of both higher and lower ALP levels with mortality in men with predialysis.[9] Another study demonstrated the presence of bone-type ALP in human vascular smooth muscle cells,[10] suggesting that increased bone metabolism activity may accelerate the development of cardiovascular events through cardiovascular calcification. Furthermore, there is growing evidence suggesting that some forms of vascular homeostasis, such as neovascularization, angiogenesis, and vascular maintenance, are closely associated with bone marrow activity,[11-13] which can be evaluated in terms of ALP activity.[1,14-17]

These findings led us to speculate that not only elevated, but also reduced, ALP levels may be associated with stroke incidence owing to the resultant impaired vascular homeostasis and vascular repair. Given that alcohol consumption might act as confounding factor for this topic, these considerations generated the hypothesis that both higher and lower levels of serum ALP are associated with risk of stroke, especially among nondrinkers. To examine this hypothesis, we analyzed the long-term follow-up data for stroke incidence among middle-aged men and women in 4 Japanese communities.

Material and Methods

The Circulatory Risk in Communities Study (CIRCS) is a prospective community-based study with the ultimate aim of preventing cardiovascular disease in Japanese populations.[18-20] A total of 12,222 persons (4822 men and 7400 women) aged 40-69 years were surveyed. Residents of the northeastern rural community of Ikawa and in the southwestern rural community of Noichi participated in this study between 1985 and 1990, residents of the central rural community of Kyowa participated between 1985 and 1991, and residents of the southwestern suburb of Yao participated between 1985 and 1994. Persons with a history of stroke or coronary heart disease (125 men and 56 women), persons with missing ALP data (578 men and 566 women) were excluded. There were no differences in cardiovascular risk factors between participants with recorded ALP data and those without these data. The remaining 10,754 participants (4098 men and 6656 women) were followed up until the end of 2004 for Kyowa and Noichi and until the end of 2007 for Ikawa and Yao to determine the incidence of stroke. The 695 persons (207 men and 488 women) who moved out of their respective communities during the follow-up and 1427 persons (843 men and 584 women) who died were censored at the date of moving out or the date of death. The median follow-up time was 16.0 years. This study was approved by the Ethics Committee of the Osaka Medical Center for Health Science and Promotion.

Baseline Examination

Details of the evaluation of cardiovascular risk factors have been described elsewhere.[20] In brief, height in stocking feet and weight in light clothing were measured, and body mass index (BMI) was calculated as weight (kg)/height $(m)^2$. Nonfasting blood samples were obtained, and serum was separated and centrifuged after blood coagulation. Serum samples were also obtained in a siliconized tube. ALP was measured with the Bessy-Lowry method, serum aspartate aminotransferase (AST) and serum alanine aminotransferase (ALT) with the Tris buffer method, and serum γ-glutamyltransferase (γ-GTP) with the γ-GTP-pNA chromogenic method. Serum total cholesterol, glucose, albumin, and creatinine were determined with the enzymatic method, hexokinase method, bromcresol green method, and noncompensated kinetic Jaffe method, respectively. SMAC (Technicon, Tarrytown, NY) was used for all measurements. In this study, ALP measurements were obtained at least twice during the baseline period for 65.7% of the participants (60.9% of the men and 68.6% of the women). The Pearson correlation coefficients for the 2 measurements were 0.72 ($P < .001$) for nondrinkers, 0.57 ($P < .001$) for drinkers, 0.59 ($P < .001$) for men, and 0.71 ($P < .001$) for women.

Glomerular filtration rate (GFR) was estimated using the established method with 3 variations recently proposed by a working group of the Japanese Chronic Kidney Disease initiative.[21] According to this adaptation, GFR $(mL/min/1.73\ m^2) = 194 \times (\text{serum creatinine (enzyme method)})^{-1.094} \times (\text{age})^{-0.287} \times (0.739 \text{ for women})$. Serum creatinine was assayed with the noncompensated kinetic Jaffe method. However, this method was recently replaced with the enzymatic method because the creatinine values established with the Jaffe method were found to be approximately 0.2 mg/dL higher than those determined with the enzymatic method owing to the presence of creatinine chromogens in the sample.[22,23] Consequently, we revised our serum data using the enzymatic method with the following formula: serum creatinine (enzymatic method) = serum creatinine (Jaffe method) − 0.2 mg/dL. Chronic kidney disease (CKD) was defined as GFR $<60\ mL/min/1.73\ m^2$ in accordance with the guidelines of the National Kidney Foundation Kidney Disease Outcomes Quality Initiative.[23]

All measurements were performed at the laboratory of the Osaka Medical Center for Health Science and Promotion, an international member of the US National Cholesterol Reference Method Laboratory Network. This laboratory has been standardized since 1975 by the Centers for Disease Control and Prevention–National Heart, Lung and Blood Institute's Lipid Standardization Program and meets the criteria for both precision and accuracy of cholesterol measurements.[24]

Physicians measured systolic and fifth-phase diastolic blood pressure in the right arm using a standard mercury