OBJECTIVE — To explore the relationship between the genetic polymorphisms of PPARγ (Pro12Ala, C1431T, and C-2821T) and the risk of ischemic stroke and to investigate whether these genetic polymorphisms of PPARγ would modify the risk of ischemic stroke among patients with hypertension or diabetes.

RESEARCH DESIGN AND METHODS — The case-control study was conducted with 537 ischemic stroke patients and 537 control subjects. A structured questionnaire was used to collect information on conventional cardiovascular risk factors and laboratory results. The genetic polymorphisms of PPARγ were determined by PCR–restriction fragment–length polymorphism.

RESULTS — A significant interaction was seen between the −2821C allele and diabetes but not between this allele and hypertension. A markedly elevated risk of ischemic stroke (odds ratio 9.7) was found in the subjects with diabetes and the −2821C allele compared with that in those without these two risk factors.

CONCLUSIONS — The −2821C allele of PPARγ was a strong predictor of ischemic stroke for diabetic patients.

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Received 15 April 2009 and accepted 27 July 2009. Published ahead of print at http://care.diabetesjournals.org/cgi/content/full/dc09-0717/DC1. Student t tests, Mann-Whitney U tests, χ² tests, and logistic regression models were used as appropriate and performed with the SAS statistical software (version 9.1). Synergy index scores were used to evaluate interaction between two risk factors (10). Pairwise linkage disequilibrium between single nucleotide polymorphism markers was evaluated by Haplovie (11).

RESULTS — The occurrences of hypertension (75.6%), diabetes (45.9%), and PPARγ C-2821C genotype (12.3%) were more common in case than in control subjects, but the distribution of Pro12Ala and C1431T was similar between the two groups. The percentage of antihypertension drugs in ischemic stroke case subjects (66.7%) was similar to that in control subjects (66.6%). Of the ischemic stroke case subjects with diabetes, 54.1% had pharmacological treatment. The percentage of pharmacological treatment for diabetes in control subjects...
Table 1—Adjusted ORs of ischemic stroke risk by hypertension, diabetes, and PPARγ−2821C allele

| Group of risk factors | Hypertension | Diabetes | PPARγ −2821C | Case/control subjects | OR (95% CI)* | Grouped OR (95% CI)* |
|----------------------|--------------|----------|-------------|-----------------------|---------------|---------------------|
| Ref.                 | −            | −        | −           | 34/93                 | 1.0           | 1.0                 |
| I                    | −            | −        | +           | 43/89                 | 1.5 (0.7–3.4) | —                   |
| II                   | −            | +        | −           | 31/15                 | 3.5 (1.2–10.0)† | —                   |
| III                  | −            | +        | +           | 101/129               | 2.7 (1.3–5.3)† | 2.3 (1.2–4.3)‡      |
| I                    | +            | −        | −           | 23/8                  | 5.3 (1.5–18.3)† | —                   |
| II                   | +            | −        | +           | 112/126               | 2.3 (1.1–4.5)† | —                   |
| III                  | +            | +        | −           | 77/43                 | 4.4 (1.8–10.4)‡ | 2.6 (1.3–5.1)‡      |
| II                   | +            | +        | +           | 115/28                | 11.6 (5.0–26.9)§ | 9.6 (4.2–21.7)§    |

Data are n unless otherwise indicated. The reference group was the study subjects without hypertension, diabetes, and the PPARγ −2821C allele. $P_{\text{trend}} < 0.0001$ among groups I–III. *Adjustment for BMI, waist circumference, history of ever smoking, dyslipidemia, and pharmacological treatment for diabetes. †$P < 0.05$. ‡$P < 0.001$. §$P = 0.0001$. +, appearance of risk factor; −, absence of risk factor.

(41.5%) was less than that in case subjects. All genetic polymorphisms were in Hardy-Weinberg equilibrium. There was a high degree of linkage disequilibrium between Pro12Ala and C-2821T.

After adjusting for BMI, waist circumference, a history of ever smoking, dyslipidemia, hypertension, diabetes, and pharmacological treatment for diabetes, the odds ratio (OR) of PPARγ C-2821C genotype was not significant. Compared with nondiabetic subjects with T-2821T genotype, an increased risk of ischemic stroke was observed in TT genotype carriers with diabetes (OR 4.2; $P = 0.07$), and the OR drastically increased to 9.7 ($P = 0.008$) in C allele carriers but not in C allele carriers without diabetes. Thus, there was a significant joint effect of the PPARγ C-2821T polymorphism and diabetes (synergy − index = 2.7) on the risk of ischemic stroke. On the other hand, no interaction between hypertension and the PPARγ C-2821T polymorphism on the risk of ischemic stroke was found (synergy − index = 0.9).

The risk of ischemic stroke was estimated for each combination of hypertension, diabetes, and the PPARγ −2821C allele, using nonhypertension, non-diabetes, and non-2821C allele carriers as the reference group (Table 1). The ORs of ischemic stroke in hypertension alone or diabetes alone were 2.7 and 3.5, respectively. Furthermore, the OR increased to 5.3 in the subjects with diabetes and the PPARγ −2821C allele, which was higher than the risk in association with hypertension or diabetes alone. The greatest OR (11.6) was seen in the subjects with hypertension, diabetes, and the PPARγ −2821C allele. A trend test indicated that the risk of ischemic stroke increased along with the accumulating number of these three risk factors ($P < 0.0001$).

**Conclusions**—Our study concluded that there was no relationship between PPARγ Pro12Ala genotype and the risk of ischemic stroke. The same result was found by Zafarmand et al. (12), but the opposite conclusion was made by Lee et al. (13). The data of genetic polymorphism of PPARγ C-2821T in diseases were scarce. This novel genetic variant was first identified in the Pima Indian population from Arizona and was reported to associate with metabolic predictors of type 2 diabetes and obesity (14). In the present study, we found a joint effect of the PPARγ −2821C allele and diabetes on the risk of ischemic stroke. Adjusting for traditional cardiovascular risk factors and pharmacological treatment for diabetes, the risk of ischemic stroke in the −2821C allele carriers with diabetes was 9.7 times greater than that of the homozygous T allele carriers without diabetes. Higher transcriptional activity was found in the −2821T allele than in the −2821C allele by using the Dual Luciferase Reporter Assay in 3T3-L1 cells (14). This implied that PPARγ could present a protective effect on ischemic stroke. Wu et al. (9) used a rat model to prove PPARγ to be a critical factor for protection against neuronal apoptosis and cerebral infarction by the mediated 14-3–3ε protein. In addition, PPARγ was reported to be an important regulator of endothelial function in the cerebral circulation, especially under conditions of high-fat–induced stress (15).

In the present study, we found that the risk of ischemic stroke in subjects with hypertension, diabetes, and the −2821C allele was 2.6 times higher than that in the subjects with hypertension and diabetes. Therefore, the combination of hypertension, diabetes, and the PPARγ −2821C allele was a strong predictor of ischemic stroke. Further studies in other populations will be very useful in establishing the contribution of this C-2821T polymorphism to ischemic stroke.

In conclusion, there was a strong interaction between the PPARγ −2821C allele and diabetes with regard to risk of ischemic stroke. Thus, the PPARγ −2821C allele was a strong predictor of ischemic stroke for diabetic patients, and PPARγ may serve as a potential target for treating ischemic stroke.

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