No association between Rho-associated coiled-coil forming protein serine/threonine kinase1 gene and schizophrenia in the Japanese population

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Rho-associated coiled-coil forming protein serine/threonine kinase (ROCK) is an important downstream effector of Rho GTPase. There are two known homolog genes of ROCK in mammals, ROCK1 and ROCK2. In the nervous system, Rho GTPase and ROCK play major roles in the development, migration, and plasticity of neurons (Van Aelst and Cline, 2004). Recently, it has been reported that genetic variability of the chimerin 2 gene, one of the Rho GTPase-activating protein, is associated with male schizophrenia in Japanese population (Hashimoto *et al.*, 2005). Moreover, ROCK1 locates at chromosome 18p that is a susceptibility locus for schizophrenia (Schwab *et al.*, 1998). Here, we attempt to perform genetic case–control study of the ROCK1 gene with schizophrenia in the Japanese population.

Three hundred and eighty schizophrenia patients [243 male and 137 female; mean age ($\pm$SD), 50.1$\pm$13.9 years], diagnosed by at least two experienced psychiatrists using *Diagnostic and Statistical Manual of Mental Disorders*, fourth edition criteria, were analyzed for this study. The following were evaluated at the time of sampling: Brief Psychiatric Rating Scale scores [mean ($\pm$SD), 39.1$\pm$10.3], the age at onset [mean ($\pm$SD), 25.3$\pm$7.5 years], and the medication equivalent to chlorpromazine [mean ($\pm$SD), 751.1$\pm$523.4 mg/day]. Seven hundred and sixty healthy control participants [429 male and 331 female; mean age ($\pm$SD), 45.5$\pm$10.8 years] were also evaluated. All participants were biologically unrelated Japanese, and resided in the neighboring area of Tokushima Prefecture in Japan. Genomic DNA was extracted from blood samples. We genotyped three single nucleotide polymorphic (SNP) markers selected from the National Center for Biotechnology Information dbSNP database. Genotyping was performed using commercially available TaqMan probes (Applied Biosystems, California, USA) for ROCK1 gene (rs8085654, rs288980, and rs1481280) with Applied Biosystems 7500 Fast Real Time PCR System (Applied Biosystems). Haplotype block structure was determined using the Haploview program (Broad Institute, Cambridge, Massachusetts, USA).

There were no significant deviations in all three SNPs from the Hardy–Weinberg equilibrium in either group. The allele frequencies in patients with schizophrenia were as follows: rs8085654A/G (0.42/0.58), rs288980C/T (0.49/0.51), and rs1481280A/C (0.32/0.68). The allele frequencies in controls were as follows: rs8085654A/G (0.43/0.57), rs288980C/T (0.49/0.51), and rs1481280A/C (0.33/0.67). The genotype frequencies in patients with schizophrenia were as follows: rs8085654, A/A(0.19), A/G(0.44), and G/G(0.36); rs288980, C/C(0.26), C/T(0.46), and T/T(0.28); rs1481280, A/A(0.11), A/C(0.41), and C/C (0.48). The genotype frequencies in controls were as follows: rs8085654, A/A(0.19), A/G(0.47), and G/G(0.34); rs288980, C/C(0.25), C/T(0.48), and T/T(0.27); rs1481280, A/A (0.11), A/C(0.44), and C/C(0.45). There were no associations between these SNPs and schizophrenia. Linkage disequilibrium among all three SNPs was high ($D' \geq 0.98$, $r^2 \geq 0.51$). With respect to haplotype block delineation, all three SNPs were in one linkage disequilibrium block. Permutation test of the three markers showed no significant difference. No correlations were detected between ROCK1 genotypes and Brief Psychiatric Rating Scale scores, amounts of antipsychotics, or age at onset. Our sample size had a post-hoc power of more than 0.8 to detect even a small effect size of W of 0.2 with α of 0.05. Accordingly, the likelihood of a type II error with our sample size seems to be considerably low. We did not find association between studied SNPs of ROCK1 and schizophrenia in the Japanese population. Further studies are needed to determine association between polymorphisms in the ROCK1 gene and schizophrenia in other populations.

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References

Hashimoto R, Yoshida M, Kunugi H, Ozaki N, Yamanouchi Y, Iwata N, *et al.* (2005). A missense polymorphism (H204R) of a Rho GTPase-activating protein, the chimerin 2 gene, is associated with schizophrenia in men. *Schizophr Res* **73**:383–385.

Schwab SG, Hallmayer J, Lerer B, Albus M, Borrmann M, Honig S, *et al.* (1998). Support for a chromosome 18p locus conferring susceptibility to functional psychoses in families with schizophrenia, by association and linkage analysis. *Am J Hum Genet* **4**:1139–1152.

Van Aelst L, Cline HT (2004). Rho GTPases and activity-dependent dendrite development. *Curr Opin Neurobiol* **14**:297–304.