

Common Variant of PDZK1, Adaptor Protein Gene of Urate Transporters, is Not Associated with Gout

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J Rheumatol 2014;41;2330-2331
<http://www.jrheum.org/content/41/11/2330>

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To the Editor:

Gout, a multifactorial disease characterized by acute inflammatory arthritis, is caused as a consequence of hyperuricemia. Previous genetic studies have revealed that gout and serum uric acid (SUA) levels have associations with various genes such as ATP-binding cassette transporter, subfamily G, member 2 (*ABCG2/BCRP*)^{1,2,3}, glucose transporter 9 (*GLUT9/SLC2A9*)¹, organic anion transporter 4 (*OAT4/SLC22A11*)^{1,4}, monocarboxylate transporter 9 (*MCT9/SLC16A9*)^{1,5}, and leukine-rich repeat-containing 16 A (*LRRC16A/CARMIL*)^{1,6}.

PDZ domain containing 1 (*PDZK1*, also known as *NHERF3*) plays a pivotal role as a scaffolding protein that forms urate transportsome^{6,7,8,9} with URAT1, ABCG2, and OAT4 (Figure 1). A single-nucleotide polymorphism (SNP), rs12129861, was first reported to have an association between *PDZK1* gene and SUA¹, which was confirmed by a replication study¹⁰. Although the minor allele of rs12129861 is shown to decrease SUA^{1,10}, to the best of our knowledge, no study to date has investigated its association with clinically defined patients with gout.

In our present study, we therefore investigated the association between clinically defined gout and rs12129861 of *PDZK1* with male Japanese subjects.

As the case group, 741 male Japanese patients with primary gout were collected from the outpatients of Midorigaoka Hospital (Osaka, Japan) and Jikei University Hospital (Tokyo, Japan). All patients were diagnosed according to the criteria established by the American College of Rheumatology¹¹. For the control group, 1302 male Japanese individuals were collected from the Japan Multi-Institutional Collaborative Cohort Study, because they had normal SUA levels (≤ 7.0 mg/dl) without any gout history. The mean ages with SD of case and control groups were 55.0 ± 13.2 and 52.7 ± 8.4 years, respectively, and their respective mean body mass index was 24.6 ± 3.5 and 23.2 ± 2.8 kg/m². Each subject participating in our study provided written informed consent. Our study was approved by the institutional ethical committees, and all procedures involved were performed in accordance with the Declaration of Helsinki. Genomic DNA was extracted from whole peripheral blood cells³. Genotyping of rs12129861 was performed by the TaqMan method (Life Technologies Corp.) with a LightCycler 480 (Roche Diagnostics)⁵. To confirm their

genotypes, more than 50 samples were subjected to direct sequencing with the following primers: forward 5'-TGT AGG TTA TTG GCA TGC C-3', and reverse 5'-CAG TAG AGA CAG GGT TTC ACC-3'. DNA sequencing analysis was performed with a 3130x1 Genetic Analyzer (Life Technologies)⁵. The chi-square test was used for association analysis with SPSS v.22.0J (IBM Japan Inc.).

Table 1 shows the genotyping result of rs12129861 for 741 patients with gout and 1302 controls. The call rate for rs12129861 was 97.4%. The p value for Hardy-Weinberg equilibrium was 0.73. A p value that suggested mistyping was not obtained. The minor allele frequencies of the variant were 0.12 and 0.13 in case and control groups, respectively, indicating that these SNP are common in both groups. The association analysis of rs12129861 showed no significant association with gout in the allele frequency mode ($p = 0.30$; Table 1). Therefore, we indicated that rs12129861, a common SNP of *PDZK1*, had no association with gout susceptibility, even though it was reported to have an association with SUA^{1,10}.

PDZK1 is a scaffolding protein which has 4 PDZ domains. Similar to another scaffolding protein Na⁺/H⁺ exchanger regulatory factor 1 (*NHERF1*), *PDZK1* is one of the key molecules for urate transportsome^{6,7,8}, and is known to bind urate transporters at its PDZ domains and to mediate the subcellular localization of those proteins (Figure 1). In addition, our previous study indicated that *LRRC16A*, which was reported to have an association with SUA¹, has an association with gout susceptibility attributable to the transportsome failure⁶. *PDZK1* is, therefore, thought to play a role in urate transport through this stabilization and/or anchoring effect of urate transporters^{7,8}. Together with the fact that *PDZK1* is expressed in the kidney⁷, it seems reasonable that the SNP of *PDZK1* would have an association with SUA levels and subsequently with gout as a result of transportsome failure.

However, our present study showed that rs12129861 of *PDZK1* has no association with gout. This may be partly because of the difference of the investigated population and/or because of the limited sample (2043 individuals). Our result with patients with gout does not deny the presence of urate transportsome involving *PDZK1* because the association between rs12129861 and SUA^{1,10}, as well as the molecular interaction among *PDZK1* and urate transporters^{6,7,8,9}, has already been reported. Nevertheless, the advantage of our study is the quality of the cases: all of the patients with gout who participated in our study were diagnosed by

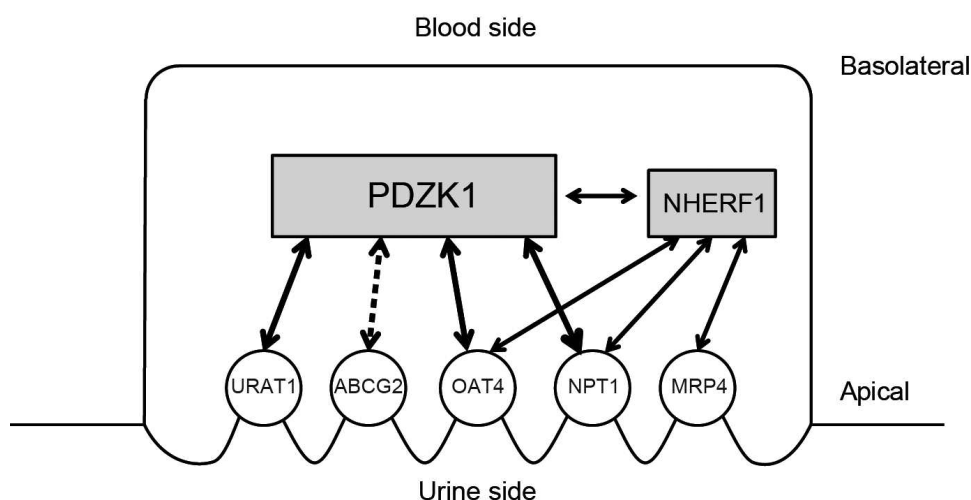


Figure 1. Urate transportsome in the renal tubular cells. *PDZK1* (also known as *NHERF3*) is a scaffolding protein that binds to several urate transporters such as URAT1, OAT4, and NPT1. As for ABCG2, the interaction with *PDZK1* is shown to be weak (dotted line)⁹. Together with *NHERF1*, which also scaffolds urate transporters (OAT4, NPT1, and MRP4), *PDZK1* plays a pivotal role in forming a urate-transporting multimolecular complex (urate transportsome) in humans.

Table 1. Association analysis of rs12129861 of *PDZK1* gene in gout cases and controls.

rs12129861	G/G	Genotypes		p	MAF	Allele Frequency Mode		95% CI
		G/A	A/A			p	OR	
Case	575	156	8	0.54	0.12	0.30	0.90	0.74–1.10
Control	951	281	19	—	0.13	—	Ref	—

MAF: minor allele frequency; Ref: reference.

expert physicians as primary gout at gout clinics. In our present study, it would be adequate to analyze the relationship between an SNP and gout susceptibility with only male patients, because female patients are rare in Japan. Indeed, only 10 female cases (1.3%) were collected at the same clinics in the same period. Therefore, the reliability of the result with our case population would be higher than those with self-reported case populations to identify the genetic factor of gout. Although further studies of *PDZK1* are necessary to reveal the relationship between *PDZK1* variants and gout, our study at least revealed that rs12129861 of *PDZK1* is not a strong genetic risk factor for gout.

Supported by grants from the Ministry of Education, Culture, Sports, Science, and Technology of Japan; the Ministry of Health, Labor, and Welfare of Japan; the Ministry of Defense of Japan; the Japan Society for the Promotion of Science; Kawano Masanori Memorial Public Interest Incorporated Foundation for Promotion of Pediatrics; the AstraZeneca VRI Research Grant; the Takeda Science Foundation; and the Gout Research Foundation of Japan.

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ACKNOWLEDGMENT

We thank all the participants involved in our study. We are indebted to Seiko Shimizu, Toshinori Chiba, Chisa Okada, Junko Abe, Keiko Gotanda, Yuki Morimoto, Naoko Katsuta, Seishiro Tatsukawa, Yuka Shichijo, Airi Akashi, Yuki Tanahashi, and Hiroki Inoue for genetic analysis. We also thank Tappei Takada, Hiroshi Nakashima, Takahiro Nakamura, Mariko Naito, Kenji Wakai, Nobuyuki Hamajima, Yukio Kato, Yoshikatsu Kanai, and Tatsuo Hosoya for helpful discussion.

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J Rheumatol 2014;41:11; doi:10.3899/jrheum.140573