Dysfunctional missense variant of OAT10/SLC22A13 decreases gout risk and serum uric acid levels

Organic anion transporter 10 (OAT10), also known as SLC22A13, has hitherto been identified as a urate transporter by *in vitro* analyses. Despite the reported expression of OAT10 on the apical membrane of the renal proximal tubular cells, the physiological impact of OAT10 on urate handling in humans remains to be elucidated. Accumulating evidence suggests that functional variants of already-characterised, physiologically
important urate transporters—URAT1/SLC22A12, GLUT9/SLC22A9, BCRP/ABCG2 and NPT1/SLC17A1—affect serum uric acid (SUA) levels and susceptibility of gout, the most common form of inflammatory arthritis. However, there are no reports on the association between OAT10 gene and either hyperuricaemia or gout. Here, for the first time, we reveal that a dysfunctional variant of OAT10 decreases both gout risk and SUA levels, suggesting OAT10 to be physiologically involved in urate reabsorption in the human kidney, as described below.

To explore exonic variants in OAT10 potentially associated with gout susceptibility, we sequenced all exons of OAT10 in 480 gout cases and 480 controls of Japanese male and conducted an association analysis (see online supplementary tables S1 and S2), followed by a replication study on 924 gout cases and 2113 controls (see online supplementary figure S1). In two identified OAT10 variants with minor allele frequency (MAF) >0.5%, only rs117371763 (c.1129C>T; p.Arg377Cys [R377C]) was significantly associated with gout susceptibility after Bonferroni correction (p=0.014). The significant association between rs117371763 and gout susceptibility was replicated, and our meta-analysis showed a significant protective effect of rs117371763 on gout susceptibility (OR=0.67; 95% CI 0.53 to 0.85; pmeta=7.8×10^{-4}) (table 1). In addition, a quantitative trait locus analysis focusing on SUA levels in 3208 individuals (see online supplementary table S3) showed that the minor allele of rs117371763 significantly decreases SUA levels (β=−0.156 mg/dL, 95% CI −0.295 to −0.018 mg/dL, p=0.027). Results were similar even after adjustment for age.

Furthermore, via a series of cell-based experiments, we identified the R377C variant as an almost null variant of OAT10 (figure 1A–C). Immunoblotting and confocal microscopic observations showed the R377C variant to have little effect on OAT10 protein levels (figure 1A) or its cellular localisation (figure 1B). Cell-based urate transport assay demonstrated that, consistent with a previous report, OAT10 wild-type can transport urate (figure 1C); however, the urate transport activity of R377C variant-expressing cells was close to that of mock cells, demonstrating that this variant disrupts OAT10’s function as a urate transporter. As it is conserved across different species (see online supplementary figure S2), R377 may be important for OAT10 function.

Considering the following three points, we conclude that OAT10 is a urate reabsorption transporter on the apical side of the renal proximal tubular cells (figure 1D). First, the R377C variant of OAT10 was almost null as a urate transporter (figure 1C). Second, this dysfunctional variant decreased SUA levels (see online supplementary table S3), suggesting that functional OAT10 is physiologically involved in a supply route of urate into the blood. Third, like URAT1/SLC22A12, which plays a pivotal role in urate transport from urine to the blood, OAT10 is reportedly expressed in the brush border membranes of the renal epithelium, therefore making it a potential target for urate-lowering therapy like URAT1. Although rs117371763 of OAT10 is common in Japanese (see online supplementary table S2), this variant is rare in other populations, including European Caucasians (see online supplementary table S4). Such populations, in which most people have functional OAT10, may offer a greater potential for OAT10 as a drug target for the treatment of gout/hyperuricaemia. Our findings will contribute to uncovering

Table 1 Association analysis of OAT10/SLC22A13 variant, rs117371763 [Arg377Cys (R377C)], with gout susceptibility

| Gout cases | Controls | p value | OR (95% CI) |
|------------|----------|---------|-------------|
| C/C | C/T | T/T | MAF (%) | C/C | C/T | T/T | MAF (%) |
| Discovery phase | | | | | | | |
| 447 | 31 | 2 | 3.65 | 427 | 46 | 6 | 6.05 | 0.014 | 0.59 (0.38 to 0.90) |
| Replication phase | | | | | | | |
| 859 | 63 | 2 | 3.63 | 1900 | 203 | 5 | 5.02 | 0.015 | 0.71 (0.53 to 0.94) |
| Meta-analysis | | | | | | | |
| 7.8×10^{-4} | 0.67 (0.53 to 0.85) |

In the meta-analysis, no apparent heterogeneity was observed (p value for Cochran’s Q test=0.48, I²=0%). MAF, minor allele frequency.
the physiological role of OAT10 as a renal urate reabsorber and its pathological involvement in urate-related disorders such as gout/hyperuricemia.

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Contributors TH, HNakaoka, YT, Tlakada and HM conceived and designed this study, TN, KH, AN, MU, TI, KI, KY, HS, NS and II assisted with research design, SS, KO, HO, TS, NS and HM collected and analysed clinical data of cases. YKawamura, SS, MU, TI, Tlamura, MN, HNakashima, MK, MT and HM collected and analysed clinical data of controls. TH, HNakaoka, SS, NS, II and HM performed genetic analysis. HNakaoka, YKawamura, HNakashima, TN and II performed statistical analyses. KM, YT, HS and Tlakada performed functional analysis. Tlakada and HM organised this collaborative study as corresponding authors. KM, TN, KH, AN, YKawai, NS, KI and KY provided intellectual input and assisted with the preparation of the manuscripts. TH, YT, YKawamura, Tlakada and HM wrote the manuscript. TH, KM, HNakaoka, YT, YKawamura and SS contributed equally to this work. All authors have read and approved the final version of the manuscript.

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