
Copyright:
The final publication is available at Cell Press via http://dx.doi.org/10.1016/j.ajhg.2016.03.026

Date deposited:
15/07/2016

Embargo release date:
19 November 2016

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Mutations in \textit{SLC26A1} Cause Nephrolithiasis

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Summary

Nephrolithiasis, a condition in which stones are present in the urinary system, affects 5~10% of individuals in their lifetime worldwide and leads to significant medical costs and morbidity. To date, mutations in more than 30 genes have been described in nephrolithiasis and these mutations explain about 15% of kidney stone cases, suggesting that additional nephrolithiasis-associated genes remain to be discovered. To identify additional genes linked to nephrolithiasis when defective, we performed targeted next-generation sequencing of hypothesized candidate genes in 348 unrelated individuals with kidney stones. We detected biallelic mutations in SLC26A1 (solute linked carried family 26 member 1) in two individuals from two unrelated families. We show by immunofluorescence, immunoblotting, and glycosylation analysis that the variant protein mimicking p.Thr185Met has defects in protein folding or trafficking. In addition, by measuring anion transporting exchange activity of SLC26A1, we demonstrate that all the identified mutations in SLC26A1 lead to decreased transporter activity. Our data identify SLC26A1 mutations as causing a recessive Mendelian form of nephrolithiasis and may have therapeutic implications.
Nephrolithiasis (MIM 167030) is a major health problem as it affects with up to 10% of the population in Western countries. Its prevalence has significantly increased among children over the last decades.\textsuperscript{1, 2} This leads to significant medical cost due to expensive surgical interventions, progression to chronic kidney disease (CKD), and additional morbidity from renal colic and secondary urinary tract infection.\textsuperscript{3} The formation of kidney stones is multifactorial resulting from an interaction of environmental, dietary, and genetic factors. Nephrolithiasis is genetically heterogenous, and mutations in at least 30 genes have been linked to this disorder.\textsuperscript{4, 5} We recently demonstrated in a cohort of 166 adults and 106 children with nephrolithiasis or nephrocalcinosis that a monogenic cause can be detected in the surprisingly high fraction of 11.4% of adult and 20.8% of childhood-onset cases.\textsuperscript{6} In a follow-up study, we detected causative mutations in 1 of 30 linked genes in 16.7% of 143 individuals who manifested with nephrolithiasis or nephrocalcinosis before 18 years of age.\textsuperscript{7} Both studies thus demonstrated a high percentage of individuals with a monogenic mutation associated with kidney stone formation. However, genetic factors have been suggested to account for nearly 50% of nephrolithiasis cases,\textsuperscript{8, 9} indicating that additional nephrolithiasis-associated gene remain to be discovered.

To identify additional genes mutated in nephrolithiasis, we generated a list of 18 hypothetical candidate genes and performed exon amplification with consecutive next-generation sequencing in a multicenter cohort of 348 individuals with nephrolithiasis or isolated nephrocalcinosis.\textsuperscript{10} The candidate genes screened in this study are listed in Table S1. We designed 210 target specific primer pairs for coding exons and the adjacent splice site of 18 genes. Amplicon sizes were chosen to range from 250 to 300 base pairs. We achieved a median sequencing coverage of 198x per individual and 192x per amplicon. Only 9/350 (2.5%) individuals had a median coverage below 20. The cohort consisted of 201 children and 147 adults who had at least one urinary stone or nephrocalcinosis, and was previously described.\textsuperscript{6, 7} For exclusion of known genetic causes of nephrolithiasis, 30 known genes previously linked to nephrolithiasis were screened by exon sequencing in these individuals, but no causative mutations were detected. Written informed consent was obtained from all individuals enrolled in this study and approval for human subjects research was obtained from the institutional review boards at the Boston Children’s Hospital.
In an individual of Macedonian descent (A3054-21) from non-consanguineous parents, who clinically presented with acute renal failure due to bilateral obstructive calculi, nephrocalcinosis and bilateral ureteropelvic junction obstruction (UPJO), we detected two compound heterozygous mutations in SLC26A1 (solute linked carried family 26 member 1; RefSeq accession number: NM_022042.3, MIM 610130) (Table 1, Figure 1 and Figure S1). This individual had recurrent episodes of renal colic due to nephrolithiasis, hypocitraturia and underwent surgery for UPJO. The two compound heterozygous missense mutations (c.554C>T;p.Thr185Met and c.1073C>T;p.Ser358Leu) in SLC26A1 are reported as a SNP in the dbSNP database, however, their minor allele frequencies are below 0.0006 and they never occurred in the homozygous state (Table 1). The amino acid residues affected by two missense mutations were conserved throughout evolution down to Danio rerio (Table 1 and Figure 1). This individual had ureteropelvic junction obstruction, therefore, we performed whole exome sequencing (WES) and examined variants in genes associated with congenital anomalies of the kidney and urinary tract (CAKUT) or UPJO. However, we could not detect any additional pathogenic variants in these genes. In addition, we examined genomic structural abnormality using WES data, but no structural variation was detected.

In a European American boy (B641-12) who had nephrolithiasis was from from consanguineous parents, we detected a homozygous missense mutation in SLC26A1 (Table 1, Figure 1 and Figure S1). The paternal grandfather of this individual also had nephrolithiasis. The detected missense mutation (c.166G>A;p.Ala56Thr) is reported as a SNP in the dbSNP database, however, its minor allele frequency is 0.0002 and it never occurred in the homozygous state (Table 1). Murine SLC26A6 has Ala at the position corresponding to amino acid position 56 of human SLC26A6, whereas the protein from Xenopus tropicalis or Danio rerio has Val, which is also chemically related as a non-charged non-polar residue (Table 1 and Figure 1).

SLC26A1 (also known as Sat1) was cloned as a sulfate transporter from liver.\textsuperscript{11} It was subsequently characterized as an anion exchanger that transports other anions including bicarbonate, chloride and oxalate, by mediating electroneutral sulfate/oxalate, sulfate/bicarbonate or oxalate/bicarbonate exchange.\textsuperscript{12, 13} The Slc26a1\textsuperscript{−/−} mouse exhibits hyposulfatemia, hypersulfaturia, calcium oxalate urolithiasis and nephrocalcinosis in the setting
of hyperoxalemia and hyperoxaluria.\(^{14}\) \(\text{Slc26a1}^{-/-}\) mice also show enhanced drug-induced liver toxicity, which may reflect reduced availability of intracellular sulfate ion for drug conjugation reactions.\(^{14}\) These findings clearly demonstrate that SLC26A1 plays a critical role in homeostasis of oxalate and sulfate in mammals, which is thought to be associated with kidney stone disease. However, to date, the contribution of genetic variants in \(\text{SLC26A1}\) to nephrolithiasis in human is unknown. Dawson \textit{et al.} examined \(\text{SLC26A1}\) in 13 individuals with recurrent calcium oxalate urolithiasis, but they were unable to not detect any biallelic mutations in \(\text{SLC26A1}\). The c.1667A>G;p.Gln556Arg variant, which was found in 6 out of 13 individuals examined with one homozygous individual, is a known SNP (rs3706622), and its allele frequency is 0.6737 (26773 homozygotes out of 79535 individuals are reported in the ExAC Browser), suggesting that this SNP is probably benign, rather than disease-causing.

The SLC26A1 transporter is known to localize at the basolateral membrane of renal proximal tubular epithelial cells and enterocytes, as well as the sinusoidal membrane of hepatocytes.\(^{14-16}\) We confirmed by immunofluorescence the localization of SLC26A1 in the rat kidney and intestine (\textbf{Figure 2A}). We show that SLC26A1 is present in the basolateral membrane of peanut lectin-positive proximal tubules in the renal cortex and basolaterally in the AQP2-positive principle cells as well as the AQP2-negative intercalated cells of collecting ducts in the medulla (\textbf{Figure 2B}). SLC26A1 also localizes to cytoplasm and basolateral membranes of ileum and colon (\textbf{Figure 2C} and \textbf{2D}).

To evaluate the impact of the identified human mutations on SLC26A1 function, we performed immunofluorescence in HEK293T cells upon overexpression of wild type (WT) and mutant SLC26A1. A mouse \(\text{Slc26a1}\) clone was purchased from Open Biosystems (clone accession BC025824) and mutant clones of \(\text{Slc26a1}\) were generated by PCR-based site-directed mutagenesis. WT protein and variant protein harboring p.Ala56Thr localized to plasma membranes (\textbf{Figure 2E}), whereas p.Thr190Met variant protein, which correspond to p.T185M of human protein, failed to reach the plasma membrane and was trapped in the endoplasmic reticulum (ER) as shown by colocalization with an ER marker, BiP (\textbf{Figure 2E}). SLC26A1 variant protein harboring p.Ser363Leu, which correspond to p.S363L of human protein, reached the cell surface, however, a significant portion of protein was trapped in the ER, suggesting a processing
defect (Figure 2E). The protein expression levels of WT and variant SLC26A1s were analyzed in HEK293T and also in PANC-1 cells (Figure 3). Because PANC-1 cells derived from pancreatic ducts retain epithelial properties and do not express endogenous SLC26A1, they constitute a suitable model for studying protein processing through transfection of plasmids encoding WT and variant SLC26A1s (Figure 3). Actually, the expression levels of p.Alα56Thr and p.Ser363Leu were reduced in in PANC-1 cells than in HEK293T cells. SLC26A1 undergoes the Golgi-mediated complex-glycosylation (band C) after an initial core-glycosylation at the ER (band B). In general, the Golgi glycosylated forms of SLC26A1 are expressed at the cell surface. When we examined the protein expression of variant proteins, we found them much reduced compared to wild type (Figure 3A and 3D). Particularly SLC26A1 p.Thr190Met was not processed to the fully glycosylated form (Figure 3A and 3D), which suggests that p.Thr190Met protein is degraded via the ER-associated protein degradation (ERAD) pathway.

To confirm whether the lower band of SLC26A1 in immunoblotting truly represents the ER form, we treated endoglycosidase H (Endo H) and peptide-N-glycosidase F (PNGase F) to the protein samples of WT and p.Thr190Met variant (Figure 3B). The band size of Endo H treated WT sample did not change, while the band size of the PNGase F treated sample decreased to the level of p.Thr190Met variant protein. The band size of p.Thr190Met variant did not change after treatment with Endo H or PNGase F. This result suggests that p.Thr190Met variant protein exists as an ER form. Real time PCR experiments were performed to determine whether the reduced protein expressions of variant proteins are due to reduced number of transcripts or to poor protein stability. We found that the number of transcripts of Slc26a1 was not significantly different in cells transfected with wild type and mutant plasmids (Figure S2), indicating that mRNA of Slc26a1 WT and mRNAs harboring mutations are not different at the transcription level. These results also indicate that reduced expression of the p.Ser363Leu variant protein is associated with deficient protein stability.

To verify functional abnormalities of SLC26A1 variant proteins, we measured the anion transporting activity of SLC26A1 by monitoring intracellular pH (pHi) in HEK293T cells after transfection with Slc26a1 plasmids (Figure 4). Measurements of pHi were performed using a pH sensitive fluorescent probe 2',7'-Bis(2-carboxyethyl)-5(6)-carboxyfluorescein acetoxymethyl
ester (BCECF-AM) as described previously. Previous studies revealed that SLC26A1 can transport chloride, bicarbonate, oxalate, and sulfate. Although SLC26A1 also has HCO$_3^-$/oxalate exchange activities (Figure S3), we rather analyzed SLC26A1-mediated SO$_4^{2-}$/HCO$_3^-$ exchange activities, because of a technical advantage of observing more robust pH$_i$ changes by HCO$_3^-$/SO$_4^{2-}$ exchange (Figure 4). HEK293T cells were firstly loaded with HCO$_3^-$ using endogenous Cl$^-$/HCO$_3^-$ exchange activities. Then the initial rate of pH$_i$ reduction as a result of HCO$_3^-$/SO$_4^{2-}$ exchange activities was analyzed after challenging the cells with 25 mM SO$_4^{2-}$. As shown in Figures 4A and 4B, cells expressing SLC26A1, but not mock-transfected cells, showed a strong HCO$_3^-$/SO$_4^{2-}$ exchange activity. Subsequently, the transport activities of SLC26A1 variant proteins were measured in the same manner. All the variants showed decreased HCO$_3^-$/SO$_4^{2-}$ exchange activities compared to wild type (Figure 4B–E). Especially the activity of the folding defect mutant, p.Thr190Met, almost vanished (Figure 4D). The summaries of difference and slope of intracellular pH are given in Figure 4F. The exchange activity of oxalate, another substrate of SLC26A1, with HCO$_3^-$ was also measured, and we observed similar but weaker activities compared with that of sulfate (Figure S3).

Taken together, using a candidate gene approach, we here demonstrate that mutations in SLC26A1 cause an autosomal-recessive form of nephrolithiasis. Our functional study indicates that all the identified mutations in SLC26A1 lead to decreased transporter activity. Slc26a1$^{-/-}$ mice have increased liver sensitivity to acetaminophen. Administration of acetaminophen led to a fourfold increase in serum alanine transaminase levels and extensive liver necrosis in Slc26a1$^{-/-}$ mice. Therefore it will be of high clinical relevance to determine whether individuals with biallelic mutations in SLC26A1 also exhibit drug-induced hepatotoxicity. Based on the findings in Slc26a1$^{-/-}$ mice, acetaminophen may be contraindicated in individuals with SLC26A1 mutations.
Acknowledgments
We thank the physicians and the participating families for their contribution. We also thank Catherine Matero, Elizabeth Andrews, Brittany Fisher, Leslie Spanes, and Jennifer Drucker for their help for patient enrollment and clinical information. F.H. is the Warren E. Grupe Professor of Pediatrics. This research was supported by grants DK1069274, DK1068306, and DK064614 from the National Institutes of Health (to F.H.), 6-FY11-241 from the March of Dimes Foundation (to F.H.), 2013R1A3A2042197 (to M.G.L.) and 2015R1D1A1A01056685 (to H. Y. G) from the National Research Foundation of Korea, Ministry of Science, ICT & Future planning, and 2015-32-0047 from the Yonsei University College Medicine (to H.Y.G).

Supplemental Data
Supplemental Data, including one table and three figures, can be found with this article online at http://www.cell.com/AJHG.

Web Resources
The URLs for data presented herein are as follows:
Ensembl Genome Browser, http://www.ensembl.org/
Mutation Taster, http://www.mutationtaster.org/
PolyPhen2, http://genetics.bwh.harvard.edu/pph2/
UCSC Genome Browser, http://genome.ucsc.edu/
REFERENCES


Figure Legends

Figure 1. Identification of recessive mutations of \textit{SLC26A1} in two individuals with nephrolithiasis.

\textbf{(A-B)} Renal sonography of individuals A3054-21 (\textbf{A}) and B641-21 (\textbf{B}) reveals kidney stones.

\textbf{(C)} Exon structure of human \textit{SLC26A1} cDNA. \textit{SLC26A1} contains three exons. Positions of start codon (ATG) and of stop codon (TAG) are indicated.

\textbf{(D)} Domain structure of SLC26A1. The transmembrane (orange) and sulphate transporter and AntiSigma factor antagonist (STAS) domains are depicted by colored bars, in relation to encoding exon position. SLC26A1 has two N-linked glycosylation sites (amino acid positions 158 and 163, red lines). Three homozygous or compound-heterozygous \textit{SLC26A1} mutations detected in two families with nephrolithiasis are shown. Family numbers (underlined), mutation, and predicted translational changes are indicated (see Table 1).

\textbf{(E)} A partial protein alignment of SLC26A1 shows evolutionary conservation of the identified missense changes.

Figure 2. Localization of SCL26A1 in kidney and intestine, and the effects of SLC26A1 mutation on subcellular localization of SLC26A1.

\textbf{(A-B)} Localization of SLC26A1 in renal cortex (\textbf{A}) and medulla (\textbf{B}). Rat kidney sections were immunostained with fluorescein isothiocyanate-labelled peanut lection (PNA) for proximal tubules, anti-SLC26A1 (rabbit polyclonal, Novus Biological, 1:200), and anti-aquaporin 2 (AQP2, mouse monoclonal, Santa Cruz Biotechnology, 1:100) for principal cells of collecting ducts antibodies. (\textbf{A}) In renal cortex SLC26A1 (red) mainly localized to proximal tubules, which were marked green by PNA. (\textbf{B}) In medulla SLC26A1 (red) was present in collecting duct principle cells (arrow heads), which were marked apically blue-green by AQP2, but also in intercalated cells (arrows), which were not marked by AQP2 (2X for inset).

\textbf{(C-D)} Localization of SLC26A1 in ileum (\textbf{C}) and colon (\textbf{D}). Intestines from rat were immunostained with anti-E-cadherin (mouse monoclonal, BD Transduction Laboratories, 1:100), anti-SLC26A1 and anti-zona occuldnes-1 (ZO-1, goat polyclonal, Santa Cruz Biotechnology, 1:100) antibodies. E-cadherin and ZO-1 mark adherens and tight junctions, respectively.
(E) Immunofluorescence of wild type (WT) and variant proteins of SLC26A1 in HEK293T cells. HEK293T cells were transfected with N-terminally Myc-tagged \textit{Slc26a1} WT of mutant clones. Cells were fixed and permeabilized with methanol, and immunostained with anti-Myc (mouse monoclonal, Cell Signaling Technology, 1:200), anti-BiP (rabbit polyclonal, Abcam, 1:100) and anti-GOLGBP1 (rabbit polyclonal, Sigma, 1:100) antibodies. BiP and GOLGBP1 mark endoplasmic reticulum and Golgi apparatus, respectively.

Figure 3. Immunoblotting and real time PCR assay of wild type and variant SLC26A1 proteins.

(A) HEK293T cells were transfected with \textit{Slc26a1} WT and mutant plasmids, and immunoblotting assays of SLC26A1 were performed. Cells were harvested and the protein samples were separated by sodium dodecyl sulfate-polyacrylamid gel electrophoresis. The separated proteins were transferred to a nitrocellulose membrane and blotted with appropriate primary and secondary antibodies. Anti-Myc (mouse monoclonal, Santa Cruz Biotechnology, 1:10000) antibody was used as primary antibody, and anti-Mouse IgG (HRP) (Thermo Scientific, Rockford, IL) antibody was used as secondary antibody. Protein bands were detected by enhanced chemiluminescence (Amersham Biosciences, Buckinghamshire, UK). Due to higher expression of wild type and p.Ala56Thr variant than p.Thr190Met or p.Ser363Leu variants, one-fifth of protein sample was used in wild type and p.Ala56Thr variant.

(B) Samples from cells expressing WT- or p.Thr190Met-SLC26A1 were digested with endoglycosidase H (Endo H, New England Biolabs, Ipswich, MA) and peptide N-glycosidase F (PNGase F, New England Biolabs, Ipswich, MA) according to the manufacturer’s instructions. In wild type, complex glycosylated form, band C, was not shifted after treatment of Endo H and was shifted to band A after treatment of PNGase F. In p.Thr190Met mutant, core glycosylated form, band B, is dominant, and the band B was shifted to band A after treatment of Endo H. p.Thr190Met mutant protein is degraded by the ER-associated degradation (ERAD) pathway.

(C) Relative expressions of band C of SLC26A1 variants in HEK293T cells. All mutants showed lower expression than wild type.

(D) Immunoblotting assays of SLC26A1 in PANC1 cells. The results were comparable with that of HEK293T cells.
(E) Treatment of glycosidase in PANC-1 cells showed similar effect with HEK293T cells.

(F) Relative expressions of Band C of SLC26A1 variants in PANC-1 cells.

**Figure 4. Measurement of HCO₃⁻/sulfate exchange activity.**

Measurements of pHᵢ in transfected HEK293T cells were performed using the pH-sensitive fluorescent probe BCECF with co-transfection of the trans-gene marker pEGFP-N1 (Life Technologies, Carlsbad, CA). BCECF fluorescence was recorded at the excitation wavelengths of 490 and 440 nm at a resolution of 2/s using a recording setup (Delta Ram; PTI Inc, Birmingham, New Jersey, USA). Removal of Cl⁻ from the bath solution induced intracellular accumulation of HCO₃⁻ (increase in pHᵢ) due to endogenous Cl⁻/HCO₃⁻ exchange activities. Then, addition of 25 mM SO₄²⁻ to the bath solution evoked a reduction in pHᵢ as a result of HCO₃⁻/SO₄²⁻ exchange activities. All experiments were performed at 37 °C.

(A-E) Representative traces of HEK 293T cells expressing EGFP alone (A), wild type (B), p.Ala56Thr (C), p.Thr190Met (D), or p.Ser358Leu (E) SLC26A1 proteins.

(F) Summaries of HCO₃⁻/sulfate exchange activity. The intrinsic buffer capacity of HEK293T cells was 10.05 mM/pH unit (at pHᵢ 7.0), and this value was not significantly different for cells transfected with WT- or mutant- Slc26a1. Therefore, the HCO₃⁻/SO₄²⁻ exchange activities were expressed as ΔpH unit/min without compensating for the buffer capacity. Data are presented as mean ± SEM. * P < 0.05; ** P < 0.01.
Table 1. Recessive *SLC26A1* mutations detected in individuals with nephrolithiasis.

<table>
<thead>
<tr>
<th>Individual</th>
<th>Sex</th>
<th>Ethnic origin</th>
<th>Parental consanguinity</th>
<th>Nucleotide change</th>
<th>Amino acid change</th>
<th>Exon (zygosity, segregation)</th>
<th>Amino acid sequence conservation</th>
<th>PP2c</th>
<th>MTd</th>
<th>Frequencies in the EVS databasee</th>
<th>Frequencies in the dbSNP databasef</th>
<th>Frequencies in the ExAC databaseg</th>
<th>Age of onset (yr)</th>
<th>Stone analysis</th>
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<tbody>
<tr>
<td>A3054-21</td>
<td>M</td>
<td>Macedonian</td>
<td>No</td>
<td>c.554C&gt;T</td>
<td>p.Thr185Met</td>
<td>3 (het, M)</td>
<td>D. rerio</td>
<td>0.996</td>
<td>DC</td>
<td>AA/GA/GG = 0/5/6480</td>
<td>rs139024319 (MAF=0.0006)</td>
<td>26/104290 (no hom)</td>
<td>5</td>
<td>CaOx</td>
</tr>
<tr>
<td>B641-12</td>
<td>M</td>
<td>European American</td>
<td>Yes</td>
<td>c.166G&gt;A</td>
<td>p.Ala56Thr</td>
<td>2 (HOM, ND)</td>
<td><em>M. musculus</em></td>
<td>0.523</td>
<td>SNP</td>
<td>TT/CT/CC = 0/5/6487</td>
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<td>59/112248 (no hom)</td>
<td>ND</td>
<td>CaOx</td>
</tr>
</tbody>
</table>

Abbreviations are as follows: CaOX, calcium oxalate; DC, disease causing; het, heterozygous in affected individual; HOM, homozygous in affected individual; M, heterozygous mutation identified in mother; MAF, minor allele frequency; MCNS, minimal change nephrotic syndrome; MI, male; MT, mutationtaster; NA, not applicable; ND, no data or DNA available; P, heterozygous mutation identified in father; PP2, PolyPhen-2 prediction score HumVar; SRNS, steroid-resistant nephrotic syndrome; SNP, single nucleotide polymorphism; yr, years.

*a* CDNA mutations are numbered according to human cDNA reference sequence NM_022042.3 (*SLC26A1*); +1 corresponds to the A of ATG translation initiation codon. *b*Amino acid residue is continually conserved throughout evolution including the species as indicated. *c*PolyPhen-2 prediction score HumVar ranges from 0 to 1.0; 0 = benign, 1.0 = probably damaging (http://genetics.bwh.harvard.edu/pph2/). *d*Mutation taster (http://www.mutationtaster.org/). *e*Exome Variant Server (http://evs.gs.washington.edu/EVS/). *f*dbSNP database (http://www.ncbi.nlm.nih.gov/SNP). *g*ExAC browser (http://exac.broadinstitute.org/).
C

D

E

[**Figure 1**]
Figure 2

PNA
SLC26A1
AQP2

E-cadherin
SLC26A1
ZO-1

WT  p.Ala56Thr  p.Thr190Met  p.Ser363Leu

BiP

GOLGBP1
Figure 3
Figure 4
Figure S1. Chromatograms of SLC26A1 mutations detected in this study. Sequence traces are shown for the mutation above the normal control. Arrow heads denote altered nucleotides.
Figure S2. Real time PCR assay of wild type and variant Slc26a1 in Panc1 cells.

Relative mRNA expressions were evaluated by real-time PCR. Total RNA was extracted from transfected Panc1 cells by using TRizol® (Invitrogen, Carlsbad, CA) according to the manufacturer’s instructions. The RNA samples were then subjected to the RT-PCR reaction with an AccuScript™ High Fidelity 1st Strand cDNA Synthesis Kit (Stratagene, La Jolla, CA) according to the manufacturer’s instructions. The primers used in the RT-PCR analysis were forward primer 5’-GAG CTG GCT GCA GAA TGT GG-3’, reverse primer 5’- GGT GTC GGT AGC GAT CTG AG-3’. The real-time PCR was performed with the StepOnePlus™ system (Applied Biosystems, Foster city, CA). Data are presented as mean ± SEM.
**Figure S3.** Measurement of HCO$_3$-/oxalate exchange activity.

pH$_i$ was measured in the same manner as Fig. 4. After removal of [Cl]$^-_o$ in HCO$_3$- buffered solution with a 5% CO$_2$ gassing, addition of 25mM oxalate makes decrease of pH$_i$.

(A-B) Representative traces of HEK 293T cells transfected with EGFP alone (A), or wild type (B) during HCO$_3$-/oxalate exchange activity measurements.
<table>
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<tr>
<th>Gene Symbol</th>
<th>Gene Name</th>
<th>Accession #</th>
<th>Associated disease</th>
<th>MIM-Phenotype #</th>
<th>Mode</th>
<th>Coding exons</th>
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<td>ADCY6</td>
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Hyperuricemic nephropathy, familial juvenile 1 / Medullary cystic kidney disease 2

AR, autosomal recessive; AD, autosomal dominant; ND, not determined; XR, X-linked dominant.