

Supplemental Information

Culture medium

- **TR-iBRB2 cells**

DMEM with 10% fetal bovine serum, 20 mM NaHCO₃, 70 mg/L benzylpenicillin, and 100 mg/L streptomycin

- **RPE-J cells**

DMEM with 4% fetal bovine serum, 20 mM NaHCO₃, 25 mM D-glucose, 0.1 mM nonessential amino acids, 60 mg/L benzylpenicillin, and 125 mg/L streptomycin

Buffer contents

- **Extracellular fluid (ECF) buffer**

122 mM NaCl, 25 mM NaHCO₃, 3 mM KCl, 1.4 mM CaCl₂, 1.2 mM MgSO₄, 0.4 mM K₂HPO₄, 10 mM D-glucose, and 10 mM HEPES, pH 7.4

- **Na⁺-free ECF buffer**

122 mM LiCl, 25 mM KHCO₃, 3 mM KCl, 1.4 mM CaCl₂, 1.2 mM MgSO₄, 0.4 mM K₂HPO₄, 10 mM D-glucose, and 10 mM HEPES, pH 7.4

- **Cl⁻-free ECF buffer**

122 mM Sodium gluconate, 25 mM NaHCO₃, 3 mM potassium gluconate, 1.4 mM calcium gluconate, 1.2 mM MgSO₄, 0.4 mM K₂HPO₄, 10 mM D-glucose, and 10 mM HEPES, pH 7.4

- **K⁺-replacement ECF buffer**

125 mM KCl, 25 mM KHCO₃, 1.4 mM CaCl₂, 1.2 mM MgSO₄, 0.4 mM K₂HPO₄, 10 mM D-glucose, and 10 mM HEPES, pH 7.4

- **Standard oocyte saline (SOS) solution**

100 mM NaCl, 2 mM KCl, 1.8 mM CaCl₂, 1 mM MgCl₂, 5 mM HEPES, 25 µg/mL gentamycin, 2.5 mM pyruvate, and 1% bovine serum albumin, pH 7.5

- **ND96 solution**

96 mM NaCl, 2mM KCl, 1mM MgCl, 1.8 mM CaCl₂, and 5 mM HEPES, pH 7.4

Statistical analysis

Statistical analyses were carried out using a one way analysis of variance (ANOVA) followed by Dunnett's test and an unpaired two-tailed Student's t-test for several and two groups, respectively. Unless otherwise indicated, all data represent means ± S.D.

Table S1. Primers for full-length cDNA cloning

Genes	Gene Bank Accession #	Orientation	Primer sequence (5' to 3')	Restriction Enzyme
rCTL1	NM_053492.3	Forward	TCTAGAATGGGCTGCTGCAGCTCCGCC	<i>Xba</i> I
		Reverse	AAGCTTTCACCTTTTCCTGAGCATCGGCTT	<i>Hind</i> III
rCTL3	NM_001013914	Forward	TCTAGATTCATCATGGGTACTCGGTGGT	<i>Xba</i> I
		Reverse	AAGCTTCTATCTCACAATGGGCCGGAGTT	<i>Hind</i> III
rCTL4	NM_212541	Forward	TCTAGATGAGCCATGGGGAAAAAGCA	<i>Xba</i> I
		Reverse	GCAGTCGTGGGTCACGATGA	<i>Not</i> I

Table S2. Primers for RT-PCR analysis

Target mRNA	Gene Bank Accession #	Orientation	Primer sequence (5' to 3')	Product size (bp)
rCTL1	NM_053492.3	Forward	CATGTGGTGGTACCACGTGGTC	162
		Reverse	CGAATAAGGCGGTTCACTGATGC	
rCTL2	NM_001134715.1	Forward	ATGTGCTCCATGCTCTACC	176
		Reverse	TTCAATGGGAAGGTCTCTGG	
rCTL3	NM_001013914	Forward	TCCAAGAATTCAAGTCACCTCACG	243
		Reverse	GACAGCAAAGCACAGGAAAAGTGT	
rCTL4	NM_212541	Forward	ATCTACCACTGCTGGCAACAGTAC	278
		Reverse	AGGAGGACGAAGGTGACCAGAG-	

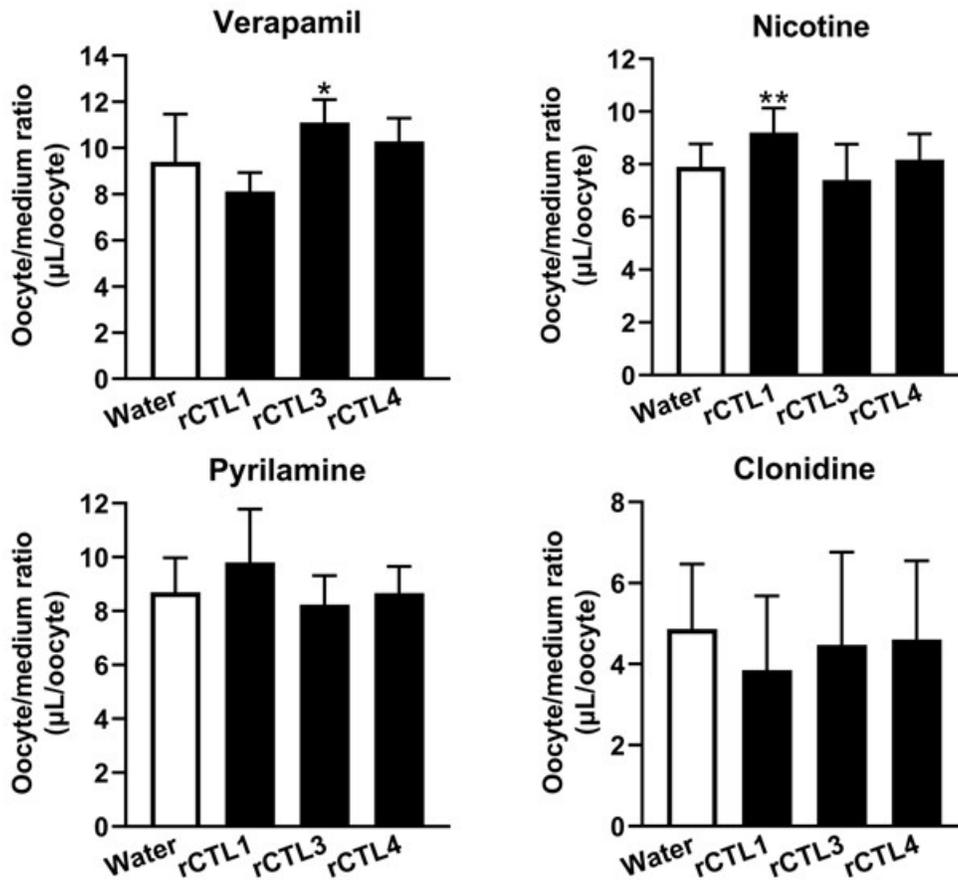


Figure S1. Uptake of cationic compounds by *Xenopus laevis* oocytes. Uptake of [³H]verapamil (0.45 μCi), [³H]nicotine (0.45 μCi), [³H]pyrilamine (0.45 μCi), and [³H]clonidine (0.45 μCi) by water-, rCTL1, rCTL3 or rCTL4 cRNA-injected oocytes was examined at 20°C for 60 min. Each column represents the mean ± S.D. (n = 9–30). **p*<0.05, ***p*<0.01, significantly different from water-injected oocytes.

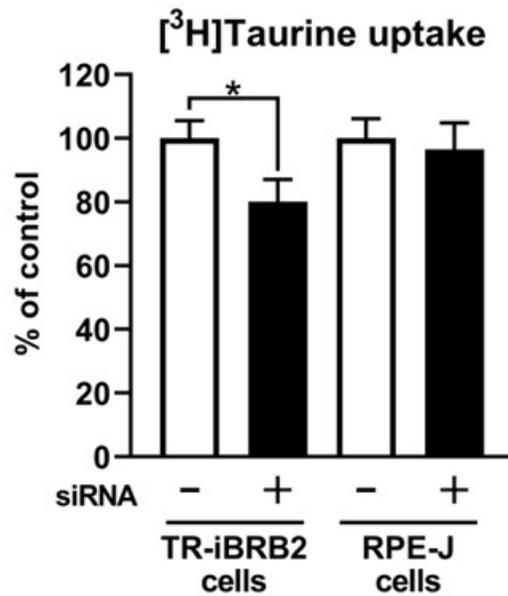


Figure S2. Effect of rCTL1 knockdown on the uptake of [³H]taurine. TR-iBRB2 cells and RPE-J cells were transfected with negative control siRNA (-) or rCTL1 siRNA (+). The uptake of [³H]taurine (0.1 μ Ci) by siRNA-transfected TR-iBRB2 cells and RPE-J cells was measured at 37°C for 10 min. Each column represents the mean \pm S.D. (n = 3). * p <0.05, significantly different from negative control siRNA-transfected cells.

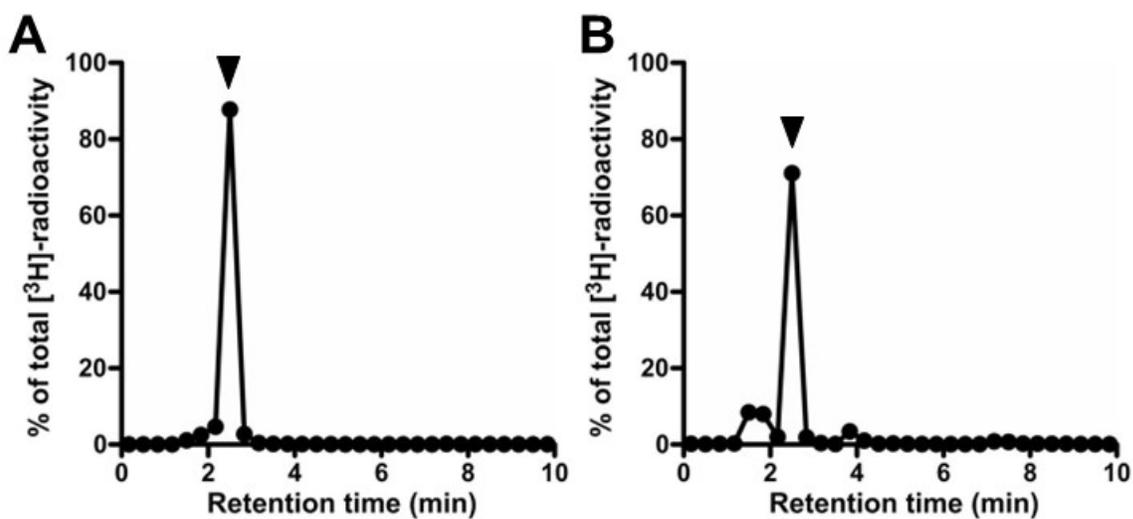


Figure S3. HPLC chromatograms of the $[^3\text{H}]$ putrescine solution (A) and tissue (retina and vitreous humor) samples (B) after the microdialysis study. Retina and vitreous humor were obtained after microdialysis study and HPLC analysis was performed at a flow rate of 1.0 mL/min with Inertsil ODS-3[®] (3 μm) (GL Sciences Inc., Tokyo, Japan) and EP-300 (Eicom, Kyoto, Japan). Mobile phase contains 121 mM sodium acetate, 7.8 mM sodium octanesulfonate, and 22% acetonitrile. The $[^3\text{H}]$ -derived radioactivity in each eluent was determined using AccuFLEX LSC7400 (Nippon RayTech Co., Ltd., Tokyo, Japan). Arrowheads indicate the retention time of $[^3\text{H}]$ putrescine.

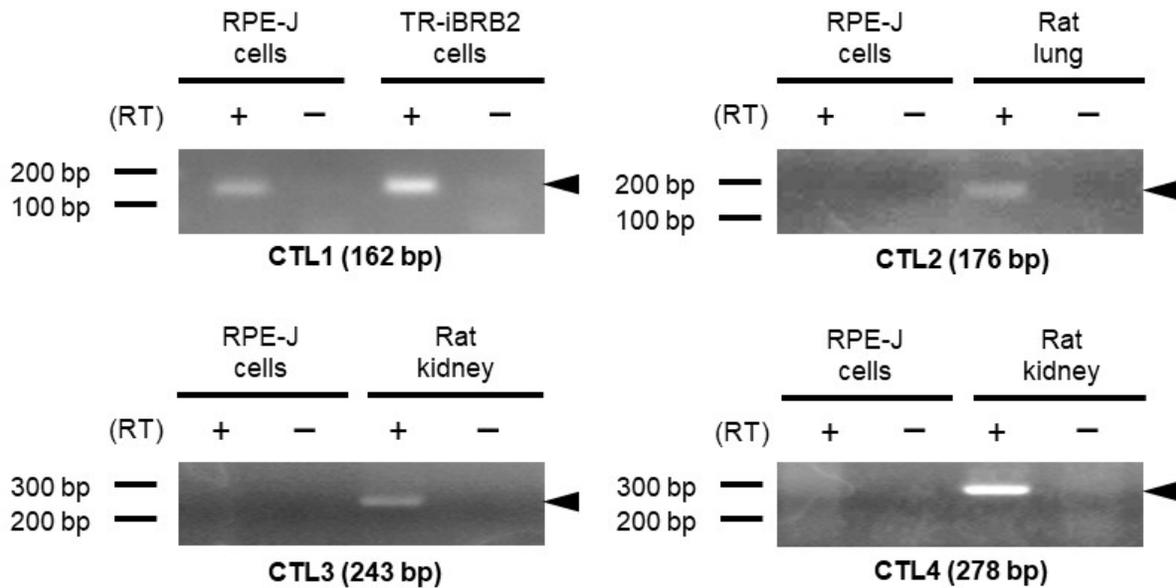


Figure S4. mRNA expression of rat CTLs in RPE-J cells. RT-PCR analysis was performed with total RNA prepared from RPE-J cells in the presence (+) or absence (-) of reverse transcriptase (RT). Total RNA prepared from TR-iBRB2 cells, rat lung, and rat kidney was used as positive control sample. Arrowhead indicates predicted product size. Primers are shown in Table S2.