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Renal Fanconi Syndrome and Hypophosphatemic Rickets in the Absence of Xenotropic and Polytropic Retroviral Receptor in the Nephron

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Abstract

Tight control of both extracellular and intracellular inorganic phosphate (Pi) levels is critical to the normal functioning of virtually all biochemical and physiological processes. The kidney participates in Pi homeostasis by controlling Pi reabsorption from the primary urine. Pi is freely filtered at the kidney glomerulus and is reabsorbed in the renal tubule by the action of the apical sodium-dependent phosphate transporters NaPi-IIa/NaPi-IIc/Pit2. However, the molecular identity of protein(s) participating in the basolateral Pi efflux remains unknown. Recent evidence has suggested that the retroviral receptor XPR1 might be involved in this process. Here we show that conditional inactivation of *Xpr1* in the renal tubule in mice results in impaired renal Pi reabsorption associated with a generalized proximal tubular dysfunction, or Fanconi syndrome, characterized by glycosuria, aminoaciduria, calciuria and albuminuria. In addition, bone histomorphometry showed that the *Xpr1*-deficient mice develop hypophosphatemic rickets secondary to renal dysfunction. Furthermore, the analysis of Pi transport in primary culture of proximal tubular cells or in freshly isolated renal tubules revealed that the Pi efflux was significantly affected in cells devoid of *Xpr1*. These results identify XPR1 as a major player in Pi homeostasis and as a potential therapeutic target in bone and kidney disorders.

SIGNIFICANCE STATEMENT

The maintenance of phosphate (Pi) homeostasis is critical for to the normal functioning of virtually all biochemical and physiological processes including preservation of bone integrity. The kidney plays a central role in Pi homeostasis by controlling urinary Pi excretion. Pi is freely filtered at the kidney glomerulus and is reabsorbed in the renal tubule by the action of the apical and basolateral Pi transporters. In this study we identified the retroviral receptor XPR1 as a new essential element of the reabsorption process. We demonstrated that mice devoid of XPR1 in the nephron exhibit renal Pi wasting associated with a generalized renal dysfunction, or Fanconi syndrome, and hypophosphatemic rickets. These results identify XPR1 as a potential therapeutic target in bone and kidney disorders.

INTRODUCTION

The xenotropic and polytropic retrovirus receptor 1 (XPR1) has long been considered as a candidate component of the Pi efflux mechanism because of its high degree of homology with PHO1 protein in plants which has been shown to mediate Pi transport from roots to shoots (1, 2). However, only recently evidence has emerged supporting a role of XPR1 in Pi transport. Battini and colleagues have shown *in vitro* that XPR1 depletion or inhibition results in a marked decrease in Pi efflux (3). Wege and Poirier have demonstrated that ectopically expressed mouse XPR1 mediates Pi efflux out of tobacco leaves (4). Most recently, Legati et al, have shown an association between genetic polymorphisms in *Xpr1* and primary familial brain calcification disorder (5). However, the role of XPR1 in the maintenance of Pi homeostasis remains unknown. Here, we addressed this issue in mice deficient for *Xpr1* in the nephron.

RESULTS AND DISCUSSION

Since *Xpr1* null mice exhibited embryonic lethality (viable pups: wild-type: 254; heterozygous: 384; null: 0), we generated mice with a doxycycline (DOX)-inducible, *Pax8*-rtTA-driven(6) conditional deletion of *Xpr1* in the renal tubule ($Xpr1^{lox/lox}$/Pax8-rtTA/LC1 mice, hereafter referred to as conditional knockout (cKO) mice). Littermate $Xpr1^{lox/lox}$ mice treated with DOX were used as controls. Males and females were investigated separately to assess possible sex differences. As shown in Supplemental Figure 1, DOX treatment resulted in a significant reduction in Xpr1 mRNA and protein levels in whole kidneys and in microdissected proximal tubules of cKO mice. The decrease in renal XPR1 expression was accompanied by a progressively increasing difference in body weights between Control and cKO mice that reached -20.4% (cKO males) and -12.1% (cKO females) 28-days after the end of DOX treatment (Figure 1A). Assessment of renal Pi handling revealed that cKO mice

exhibit hypophosphatemia (Figure 1B), phosphaturia (transient in males, Supplemental Figure 2), inappropriately low maximal tubular reabsorption of Pi per volume of glomerular filtrate (TmPi/GFR) (Figure 1C) and significantly increased fractional excretion of Pi (Figure 1D). Furthermore, we assessed the role of XPR1 in Pi efflux in primary cultures of proximal tubular cells isolated from kidneys of DOX-untreated Control and cKO mice. Xpr1 deficiency was induced *ex vivo* by 24-hours DOX exposure. Twenty-four hours after the end of DOX treatment the Xpr1 mRNA expression was significantly decreased in the proximal tubular cells isolated from cKO mice, as assessed by qPCR (Xpr1 mRNA expression in cKO cells vs. Control cells: 18.9±7.3 %; n=3; p=0.008, t-test). As shown in Figure 1E, proximal tubular cells from cKO mice had a non-significant trend towards lower [^{33}Pi]phosphate uptake. In contrast, [^{33}P]phosphate efflux was strongly affected by XPR1 deficiency (Figure 1F). The latter correlated with higher percentage of [^{33}P]phosphate remaining in the proximal tubular cells from cKO mice after 60 min of efflux (Figure 1G). Importantly, efflux of [^{14}C]glucose was not different between proximal tubular cells isolated from kidneys of Control or cKO mice (Figure 1H), indicating that the short-term *ex-vivo* Xpr1 deficiency did not result in the overall depression of efflux transport activity. The Pi efflux was also evaluated in renal tubules freshly isolated from kidneys of Control or cKO mice treated with DOX for 5 days. As shown in Figure 1I, the Pi uptake was similar in both genotypes. Importantly, the uptake was significantly reduced in the presence of phosphonoformic acid (PFA), a competitive inhibitor of apical Na^+/Pi co-transport. The efflux kinetics of Pi, which was measured in the presence of PFA, was markedly slowed in tubules isolated from kidneys of cKO mice, thus providing additional evidence for a role of XPR1 in Pi export (Figure 1I).

Analysis of urine samples revealed that 1 week after the beginning of DOX treatment cKO mice develop generalized proximal tubule dysfunction, or renal Fanconi syndrome, characterized by aminoaciduria (Figure 2A), glycosuria (Figure 2B), albuminuria (Figure 2C),

magnesuria (Supplemental Figure 3A), calciuria (Supplemental Figure 3B), lower urinary pH (Supplemental Figure 4A), polyuria (Supplemental Figure 4B) and decreased urine osmolality (Supplemental Figure 4C). Transcriptome analysis of kidneys from Control and cKO mice (males) revealed dramatic changes in expression levels of RNAs encoding proteins involved in apical Pi reabsorption (NaPi-IIa (Slc34a1): -7.19-fold; NaPi-IIc (Slc34a3): -25.37-fold), in glucose reabsorption (SGLT2 (Slc5a2): -2.88-fold; GLUT2 (Slc2a2): -2.87-fold); in amino acid transport (LAT2 (Slc7a8): -4.59-fold; BAT1 (Slc7a9): -4.24-fold; LAT1 (Slc7a7): -3.36-fold; 4F2hc (Slc3a2): -1.83-fold), and in endocytic receptors required for re-uptake of filtered albumin in the proximal tubule (megalin (Lrp2): -3.52-fold; cubilin (Cubn): -3.40-fold) (Supplemental Table 1). The impairment in tubular albumin reabsorption was assessed functionally by confocal microscopy analysis of kidney slices prepared from kidneys of mice intravenously injected with fluorescent albumin (Texas Red- or TR-albumin). As shown in Figure 2D, TR-albumin was abundantly present in the subapical region of the proximal tubular cells in kidneys of Control mice, while the TR fluorescence intensity was significantly lower in kidneys of cKO mice.

The kidneys of cKO mice exhibited reduced expression of genes encoding mitochondrially located proteins (Supplemental Figures 5A and 5B) despite normal mitochondrial biogenesis (Supplemental Figure 5C) and apparently normal mitochondria, as examined by electron microscopy (Supplemental Figure 5E). The NAD^+/NADH ratio was significantly reduced in kidneys of cKO mice, suggesting a shift in the metabolic status resulting from the XPR1 deficiency (Supplemental Figure 5D).

The glomerular filtration rate (GFR) was significantly decreased in male cKO mice along with an increase in plasma creatinine levels in cKO mice of both sexes (Table). The cKO mice exhibited slightly higher calcemia whereas plasma levels of glucose, sodium and potassium and plasma osmolality were not different from Controls (Table). Plasma

aldosterone levels were significantly increased suggesting extracellular volume contraction in cKO mice (Table).

Hypophosphatemia and decreased TmPi/GFR prompted us to study the bone phenotype in cKO mice. Analysis of vertebrae by micro-computed tomography (μCT) revealed severely decreased bone mineral density (BMD), bone volume per total volume (BV/TV), trabecular thickness (Tb.Th) and trabecular number (Tb.N) in male cKO mice and similar but milder features in female cKO mice (Figure 3A and Supplemental Table 2). The μCT analysis of femora showed significantly decreased thickness and tissue mineral density (TMD) in the distal dia- and metaphyseal cortical bone in male cKO mice, with similar but non-significant changes in female cKO mice, and largely unaffected trabecular bone of distal metaphysis (Supplemental Table 3).

Vertebral specimens of male Control and cKO mice were further analyzed by non-decalcified bone histomorphometry. Although not clearly visible at low magnification (Figure 3B) high magnification analysis showed a striking increase in all unmineralized osteoid parameters in cKO mice (Supplemental Table 4). The excessive osteoid in vertebrae of cKO mice is distinctly visible on a representative image of Toluidine Blue staining (Figure 3C). Cellular osteoblast parameters (the number of osteoblasts (N.Ob/B.Pm) and the osteoblast surface (Ob.S/B.Pm)) were unchanged in cKO mice, whereas the number of osteoclasts (N.Oc/B.Pm) was increased (Supplemental Table 4). Collectively, these data revealed a highly excessive fraction of unmineralized bone in cKO mice consistent with rickets.

To gain further insight into the molecular mechanisms underlying the defective bone mineralization in cKO mice, we measured plasma levels of hormones involved in calcium/phosphate homeostasis and bone turnover biomarkers (Table). Most strikingly, fibroblast growth factor 23 (FGF23) levels were undetectable in cKO mice of both sexes, while $1,25(OH)_2$-D_3 (Calcitriol) and parathyroid hormone (PTH) levels were unchanged.

Collagen degradation product CTX1 was significantly increased in male cKO mice, and a non-significant trend in the same direction was found in female cKO mice, suggesting an increase in bone resorption consistent with the increased osteoclast numbers observed. However, alkaline phosphatase (ALP) activity was unchanged. The levels of the osteoblast-produced hormone osteocalcin were increased in male cKO mice. To summarize, distinct signs of overall altered bone turnover were present in cKO mice.

To conclude, mice deficient for *Xpr1* in the renal tubule develop complete Fanconi syndrome and hypophosphatemic rickets. The severity of renal dysfunction was similar in cKO mice of both sexes, whereas the bone phenotype was more prominent in males compared to females, an observation that has been made in human patients(7). Hypophosphatemic ricket represents a heterogeneous entity that can be further divided into conditions associated with high FGF23 levels and suppressed $1,25(OH)_2$-D_3 such as X-linked hypophosphatemic rickets (XLH), autosomal recessive hypophosphatemic rickets (ARHR) or with low FGF23 and high $1,25(OH)_2$-D_3 levels, found when defects of renal phosphate transport are present. Indeed, mutations of NaPi-IIa and NaPi-IIc, the two sodium-phosphate co-transporters present in the brush border of the proximal tubule lead to hereditary hypophosphatemic rickets with hypercalciuria (HHRH) (8, 9). Here, we provide evidence for involvement of XPR1 in hypophosphatemic rickets associated with low FGF23 levels and normal $1,25(OH)_2$-vitamin D levels reminiscent of HHRH. Furthermore, we show that renal XPR1 is essential for phosphate homeostasis and bone physiology and open new avenues for treatment options.

METHODS

Detailed methods are described in the Supplemental Methods.

ACKNOWLEDGMENTS

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Figure legends.

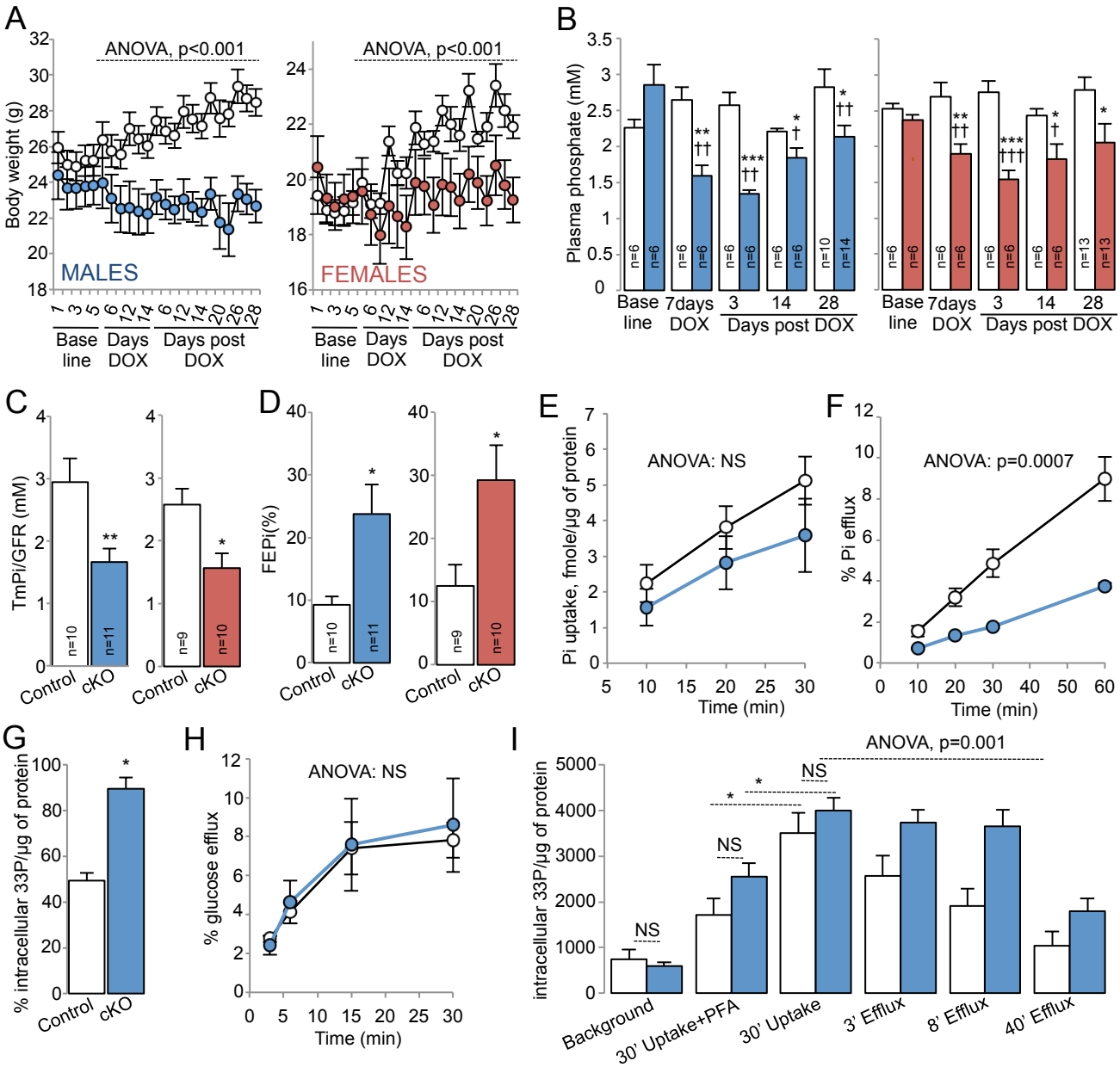
Figure 1. Altered renal handling of Pi in cKO mice. White circles/bars indicate Controls mice. Blue and red circles/bars indicate male or female cKO mice, respectively. **A.** Body weight in Control and cKO male (left panel) or female (right panel) mice. The body weight was measured during 5 days preceding DOX treatment (Baseline), during 2-weeks period of DOX treatment (Days DOX) and, during 28 days following DOX withdrawal (Days post DOX). n=6 in each group; ANOVA. **B.** Plasma Pi levels in Control and cKO male (left panel) or female (right panel) mice. Plasma Pi concentration was measured on the day preceding the 2-week period of DOX treatment (Baseline), on day 7 of DOX treatment (7 days DOX) and, on days 3, 14, 21 and 28 following DOX withdrawal (Days post DOX). * - indicates statistical significance between Control and cKO mice. *p<0.05; **p<0.01; ***p<0.001; t-test. † - indicates statistical significance between plasma Pi levels measured at baseline and plasma Pi levels measured on day 7 of DOX treatment or after DOX withdrawal (days 3, 14 and 28). †p<0.05; ††p<0.01; †††p<0.001; t-test. **C.** TmPi/GFR in Control and cKO male (left panel) or female (right panel) mice. The TmPi/GFR was determined on day 28 following DOX withdrawal. *p<0.05; **p<0.01; t-test. **D.** FEPi in Control and cKO male (left panel) or female (right panel) mice. The FEPi was determined on day 28 following DOX withdrawal. *p<0.05; t-test. **E.** [^{33}Pi]phosphate uptake in primary cultures of proximal tubule cells isolated from DOX-untreated Control or male cKO mice. Cells were exposed to DOX for 24 hours and the [^{33}Pi]phosphate uptake was measured 24-hours after the end of DOX treatment (see Supplementary Methods for details). n=4 in each group; ANOVA. **F.** [^{33}P]phosphate efflux from primary cultures of Control or cKO proximal tubule cells. n=4 in each group; ANOVA. **G.** [^{33}P]phosphate remaining in primary cultures of Control or cKO proximal tubule cells at the end of the efflux experiment (60 min of efflux); n=4 in each group. *p<0.05; t-test. **H.** [^{14}C]glucose efflux from primary cultures of Control or cKO

proximal tubule cells. n=4 in each group; ANOVA. **I.** [^{33}P]phosphate uptake (30 min) and efflux (3 min, 8 min and 40 min) from renal tubules freshly isolated from kidney of Control or cKO mice induced with DOX for 5 days. Pi uptake was determined in the presence or absence of 5 mMol PFA. Pi efflux was measured in the presence of 5 mMol PFA (see Supplementary Methods for details). Background represents nonspecific binding of [^{33}P]phosphate to the renal tubules. n=4 in each group. The difference in the efflux kinetics was evaluated by ANOVA (genotype-time interaction). The differences in Background, 30' uptake + PFA and 30' uptake conditions was evaluated by *t*-test. **p*<0.05. NS – non significant. Numbers inside of bars represent the number of animals. Data are mean±SEM.

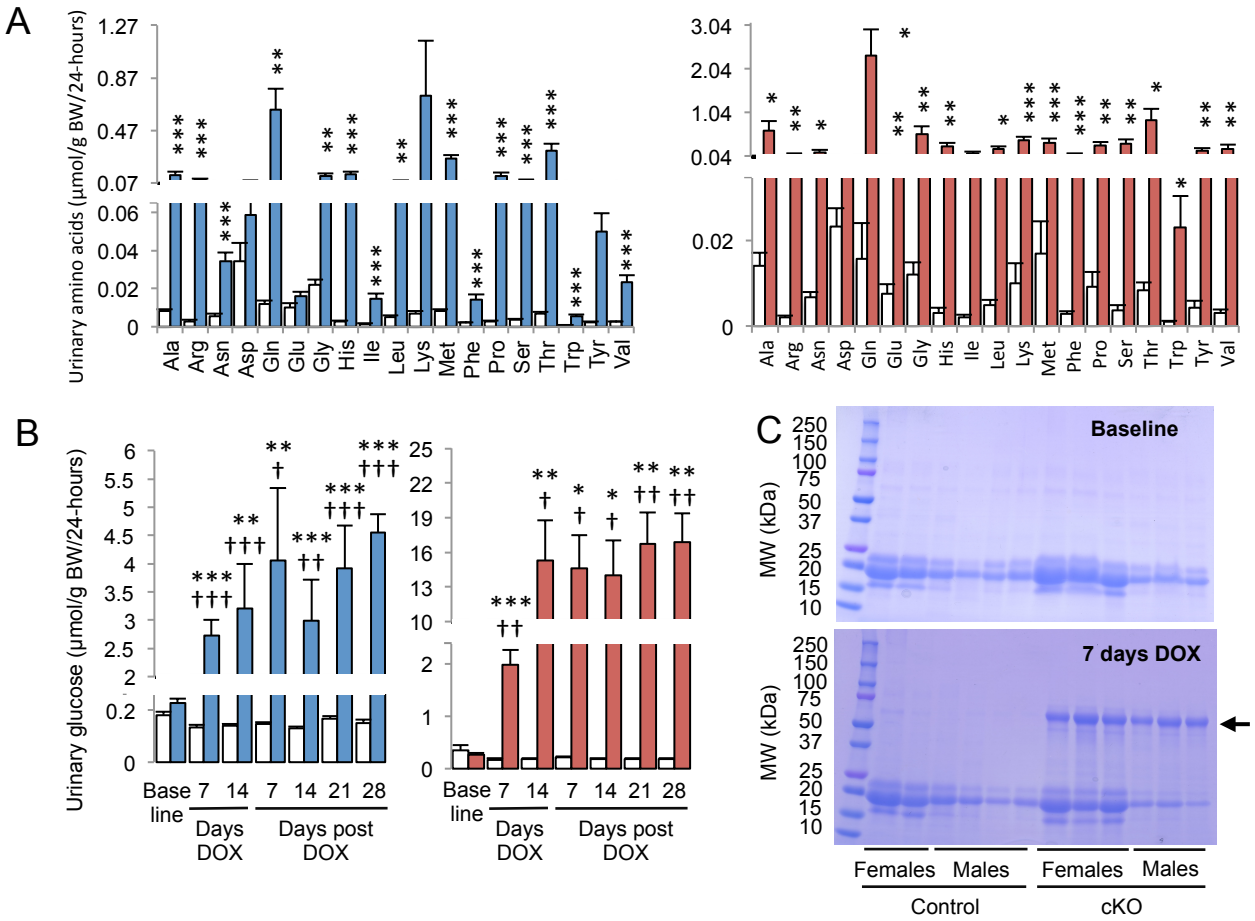
Figure 2. XPR1 deficiency in the nephron is associated with aminoaciduria, glucosuria, albuminuria and impaired albumin reabsorption in the proximal tubule. **A.** Aminoaciduria in cKO mice. The urinary excretion rate of 19 out of 20 proteinogenic amino acids (at the exception of cysteine) was measured by mass spectrometry on urines collected on day 28 following DOX withdrawal. White bars indicate the urinary excretion rates of amino acids in Control mice. Blue and red bars indicate the urinary excretion rates of amino acids in male or female cKO mice, respectively; n=6 in each group. **p*<0.05; ***p*<0.01; ****p*<0.001; *t*-test. **B.** Glucosuria in cKO mice. The urinary excretion rate of glucose was measured on urines collected on the day preceding the 2-week period of DOX treatment (Baseline), on days 7 and 14 of DOX treatment (Days DOX) and, on days 7, 14, 21 and 28 following DOX withdrawal (Days post DOX). White bars indicate the urinary excretion rates of glucose in Control mice (n=6 for males and n=4 for females). Blue and red bars indicate the urinary excretion rates of glucose in male or female cKO mice, respectively (n=6 for males and n=6 for females). * - indicates statistical significance between Control and cKO mice. **p*<0.05; ***p*<0.01; ****p*<0.001; *t*-test. † - indicates statistical significance between the urinary excretion rates of glucose measured at baseline and the urinary excretion rates of glucose measured during the

period of DOX treatment or after DOX withdrawal. †p<0.05; ††p<0.01; †††p<0.001; t-test. **C.** Albuminuria associated with XPR1 deficiency. Urine (5 μl) was run on SDS-PAGE and stained with Coomassie blue. Urine was collected from the same mice on the day preceding the 2 weeks DOX treatment period (Baseline) or on day 7 of DOX treatment (7 days DOX). The albumin band (67 kDa) is indicated by an arrow. **D.** Decreased tubular reabsorption of Texas Red – (TR)-albumin in kidneys of cKO mice. Confocal microscopy analysis of kidney slices prepared from perfusion-fixed kidneys of TR albumin-injected Control (left panel) or cKO (right panel) mice. Mice were sacrificed 5 minutes after intravenous injection of TR-albumin. Data are mean±SEM.

Figure 3. XPR1 deficiency in the nephron causes vertebral osteomalacia in male mice. **A**. 3-dimensional reconstructions of 400 μm thick coronal sections of L5 vertebral bodies scanned by mCT revealed an impaired trabecular network in male cKO mice (images representative of 2 Control and 3 cKO mice). **B.** Calcium staining of vertebral sections by von Kossa revealed no significant change in trabecular bone of cKO animals. **C.** Toluidine Blue staining of vertebral bone revealed a prominent osteoidosis (osteoid seam in light blue) in cKO mice. **B, C**: Representative images of 5 Control and 5 cKO mice; 4 μm thick sections of non-decalcified bone viewed under 2X (**B**) or 20X (**C**) magnification; scale bars 500 μm (**B**) and 50 μm (**C**). Mice were sacrificed on day 28 following DOX withdrawal.



Ansermet et al. Figure 1.



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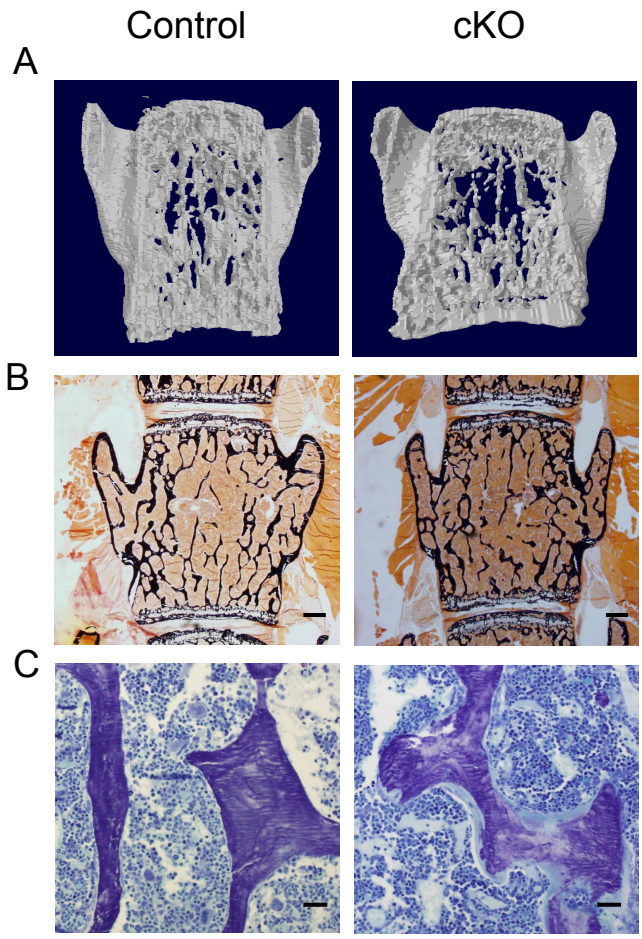
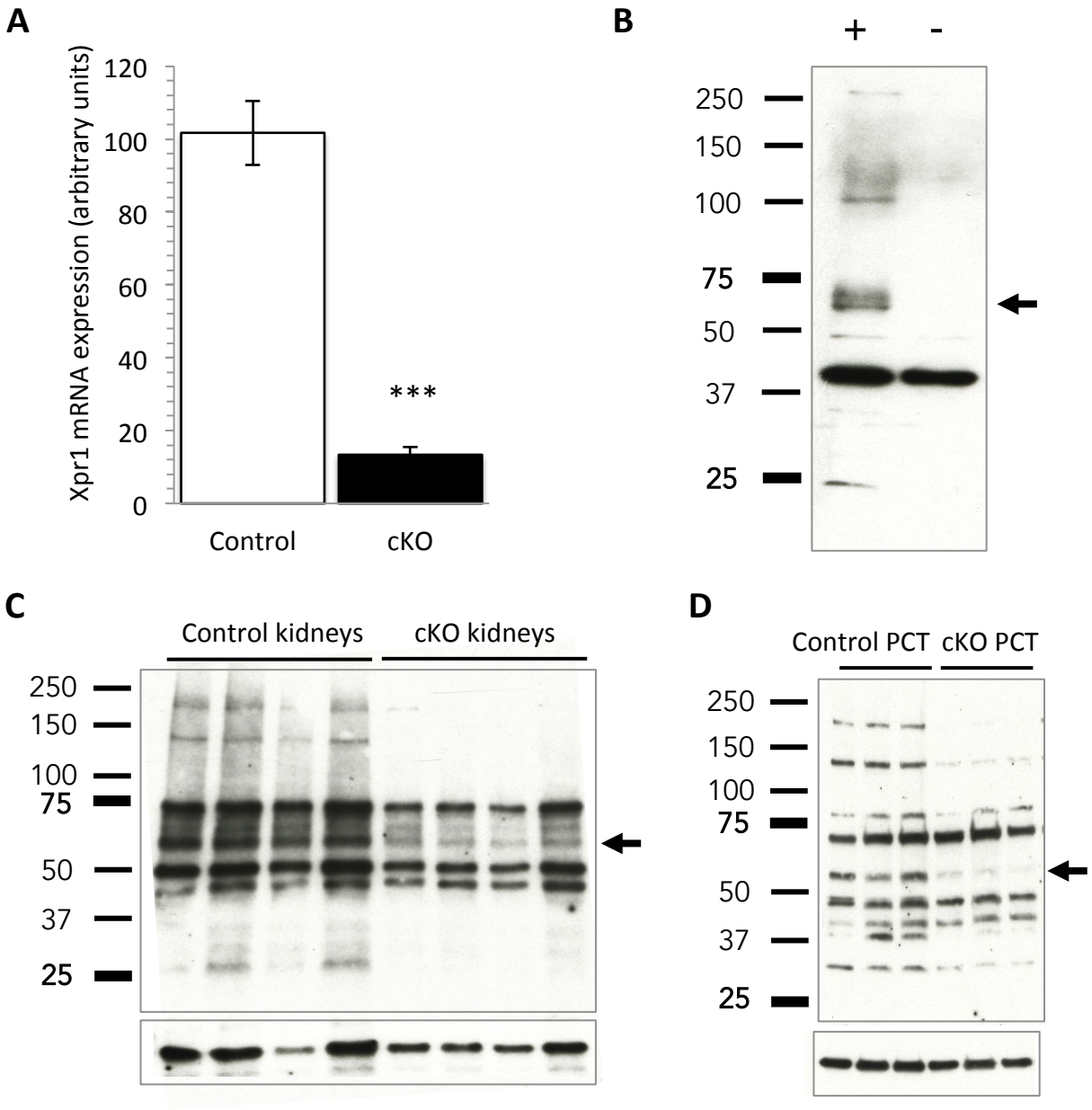


Ansermet et al. Figure 3.

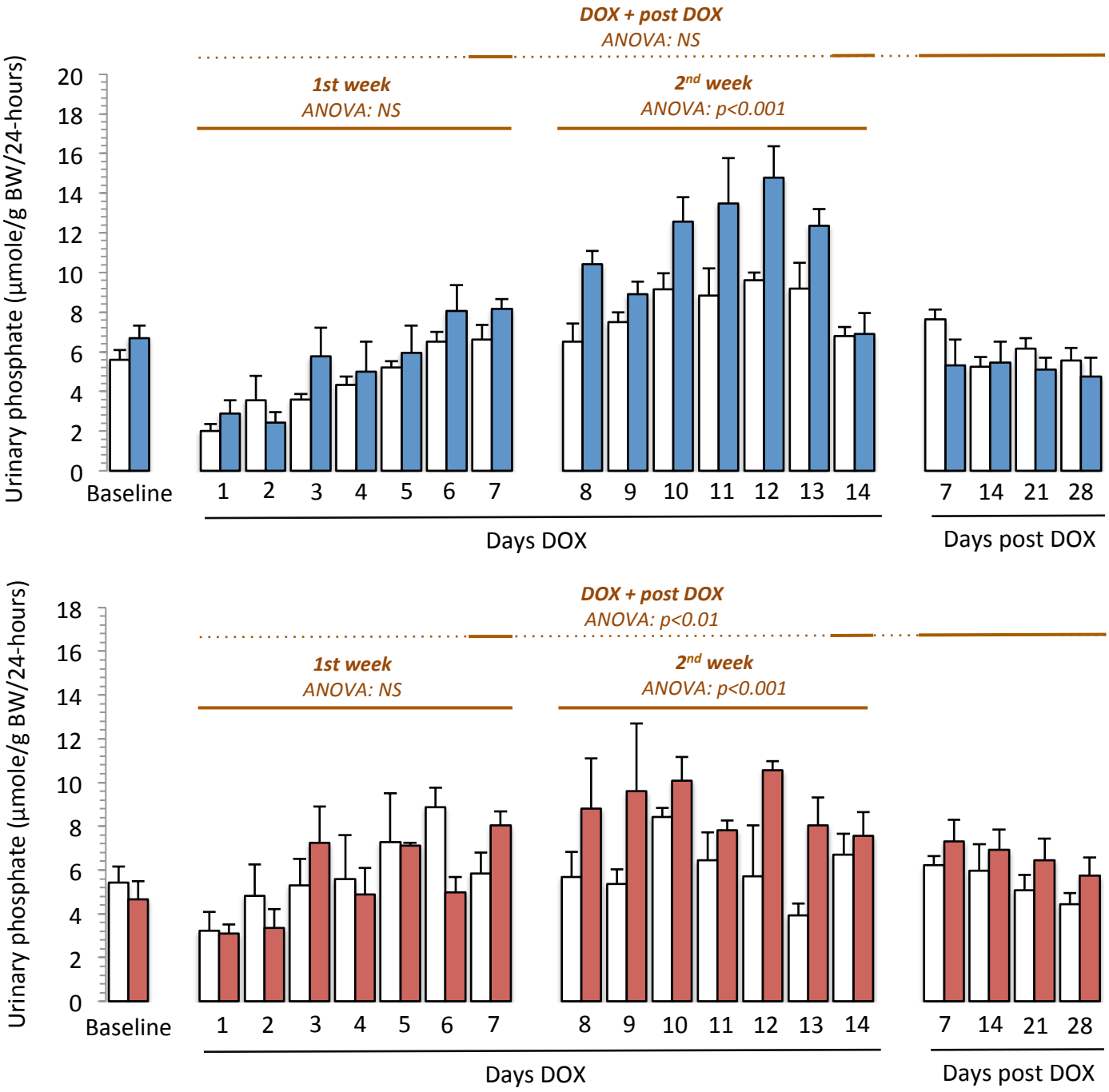
Table. Plasma chemistry and GFR in Control and cKO mice sacrificed on day 28 following Dox withdrawal.

	Males			Females		
	Control	cKO	p	Control	cKO	p
PLASMA						
Osmolality, mosm/kg H2O	325.6±1.1 (6)	324.6±5.6 (6)	0.86	324.4±2.2 (4)	323.8±2.1 (6)	0.86
Ca2+, mM	2.12±0.01 (4)	2.18±0.02 (5)	0.029	2.11±0.02 (5)	2.41±0.06 (4)	0.001
Na+, mM	156.5±2.2 (6)	159.9±4.2 (6)	0.44	158.6±1.2 (5)	160.9±2.89 (4)	0.58
K+, mM	4.04±0.16 (6)	3.64±0.20 (3)	0.16	3.74±0.29 (5)	3.19±0.09 (4)	0.09
Creatinine, μM	11.03±1.03 (16)	21.24±1.68 (15)	1.26E-05	14.03±1.69 (16)	20.16±1.37 (16)	0.008
GFR(inulin), μl/min	235±13 (5)	147±24 (5)	0.012			
aldosterone, pg/ml	202.46±65.27 (8)	408.25±56.76 (8)	0.032	222.02±54.06 (9)	506.85±78.97 (8)	0.008
FGF23, pg/ml	122.95±15.57 (5)	<30 (4)	0.0002	142.61±20.37 (4)	<30 (5)	9.33E-05
CTX-1, pg/ml	5.95±0.87 (4)	17.71±2.34 (4)	0.003	6.56±0.30 (4)	30.31±12.85 (4)	0.11
1.25(OH)2-D3 nmole/ml	88.97±13.41 (5)	66.04±5.97 (5)	0.16	56.53±4.57 (5)	55.86±6.13 (5)	0.93
PTH, pg/ml	20.29±7.30 (6)	34.05±11.34 (6)	0.33	39.25±12.75 (6)	15.11±6.83 (6)	0.12
ALP activity, U/l	3.87±1.81 (4)	3.77±0.75 (4)	0.96	9.06±4.43 (4)	7.48±3.45 (4)	0.79
TRAP, ng/ml	124.13±13.02 (4)	92.54±12.13 (4)	0.13	98.07±11.79 (4)	62.12±6.89 (4)	0.03
Osteocalcin, ng/ml	66.13±18.61 (6)	210.79±59.18 (4)	0.03	104.59±23.77 (4)	145.92±14.79 (3)	0.23

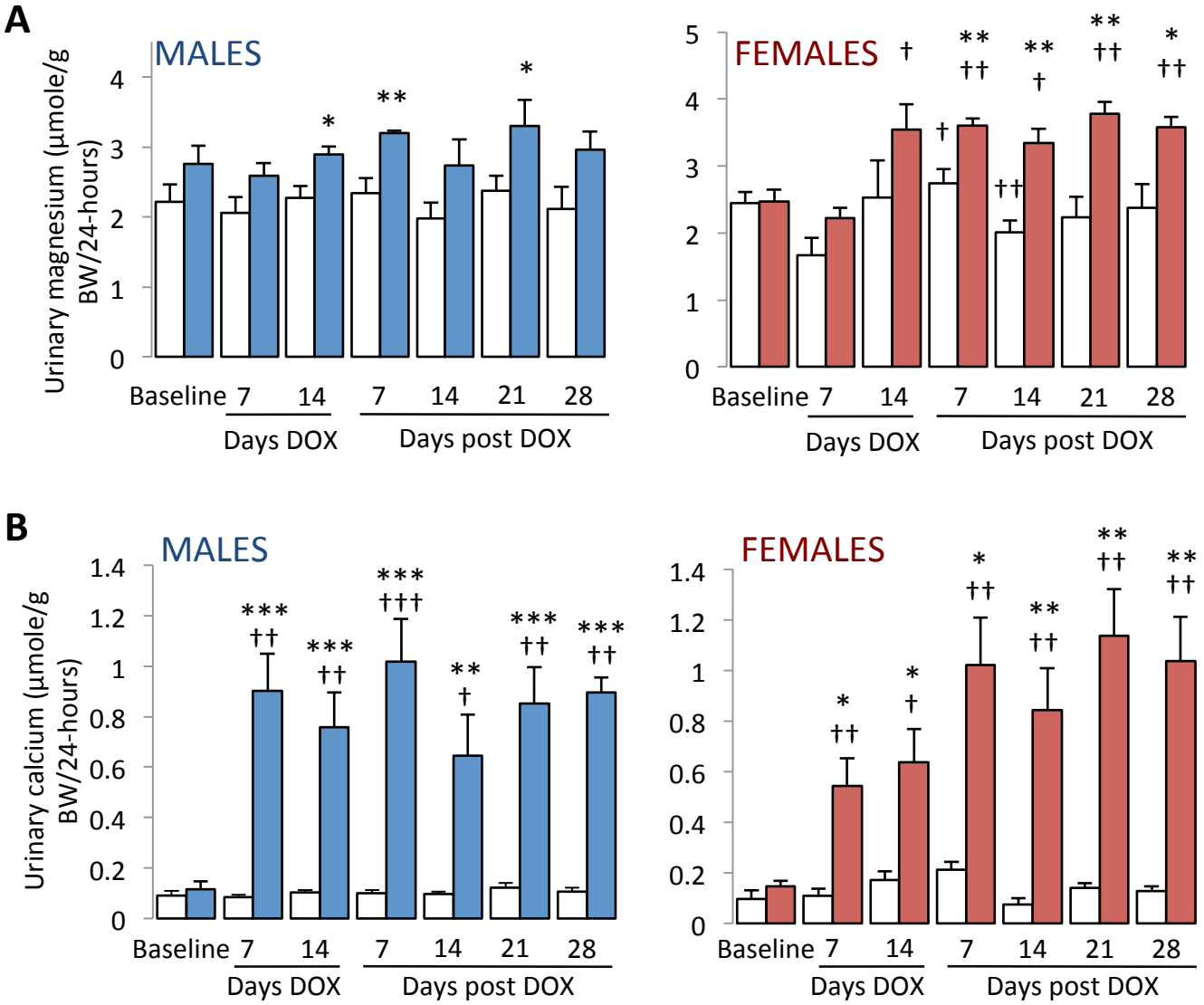
Data are means±SEM. Unpaired *t*-test.



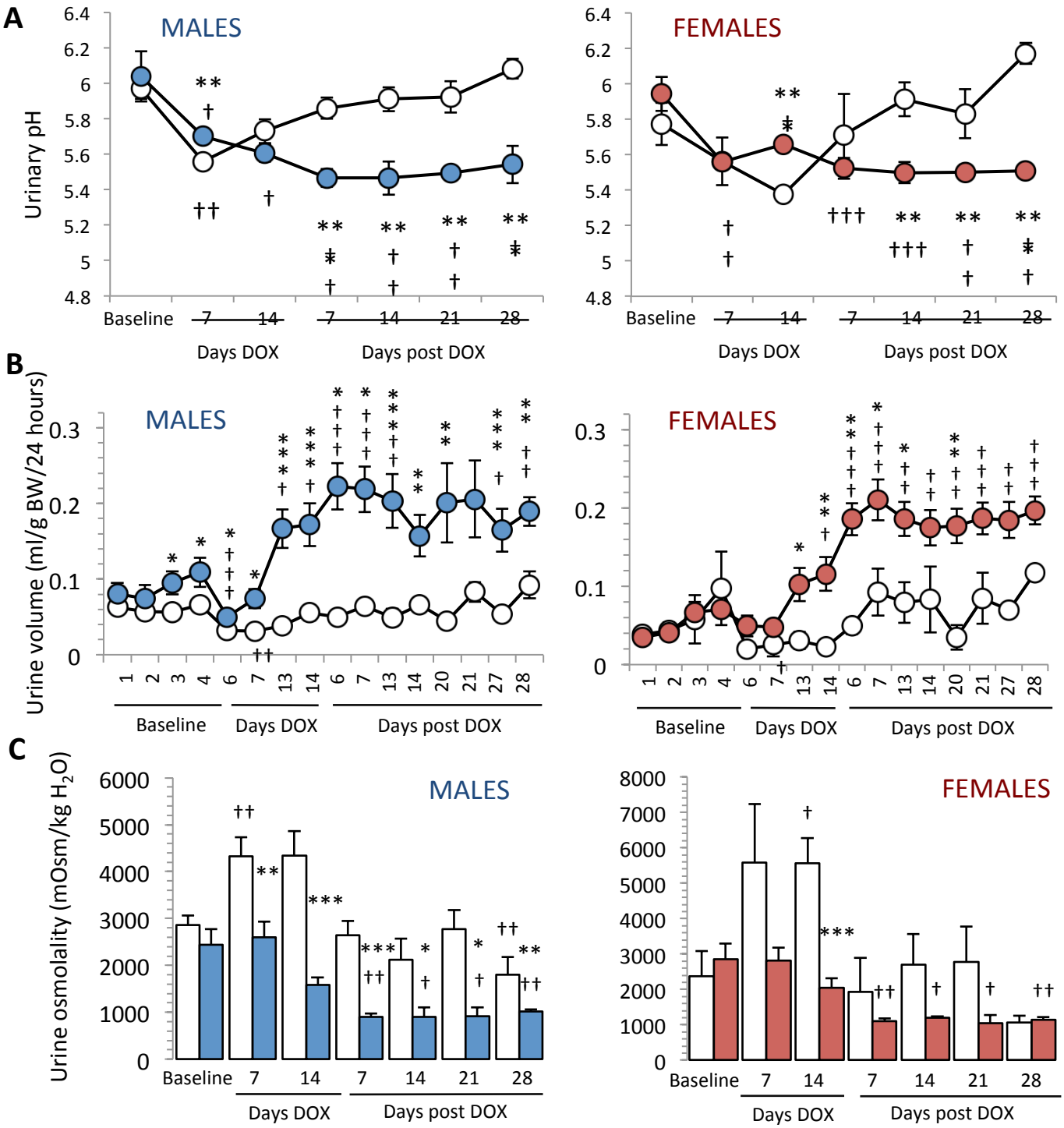
Supplemental Figure 1. Xpr1 mRNA and protein levels are significantly reduced in kidneys of cKO mice. **A.** qPCR analysis of Xpr1 mRNA expression levels in kidneys of Control and cKO male mice (arbitrary units). The mice were sacrificed on day 28 following DOX withdrawal. n=6 for both genotypes. ***p<0.001, unpaired t-test. **B.** The specificity of anti-N-term-XPR1 antibody from Proteintech (Manchester, UK) was tested by Western blotting of protein extracts prepared from mouse Xpr1 cRNA-injected (+) or water-injected (-) *Xenopus laevis* oocytes. The most prominent specific band was observed at ~ 60 kDa. Since the predicted molecular weight for XPR1 is ~ 80 kDa, these data suggest proteolytic cleavage at the C-terminus. **C.** Western blotting of membrane protein exctracts prepared from kidneys of Control and cKO mice sacrificed on day 28 following DOX withdrawal. n=4 for both genotypes. The ~ 60 kDa protein band recognized by the anti-N-term-XPR1 antibody in Control mice had significantly reduced intensity in cKO mice. **D.** Western blotting of protein extracts prepared from the proximal convoluted tubules (PCT) microdissected from kidneys of Control or cKO mice sacrificed on day 28 following DOX withdrawal. n=3 for both genotypes. The ~ 60 kDa protein band recognized by the anti-N-term-XPR1 antibody in Control mice had significantly reduced intensity in cKO mice.



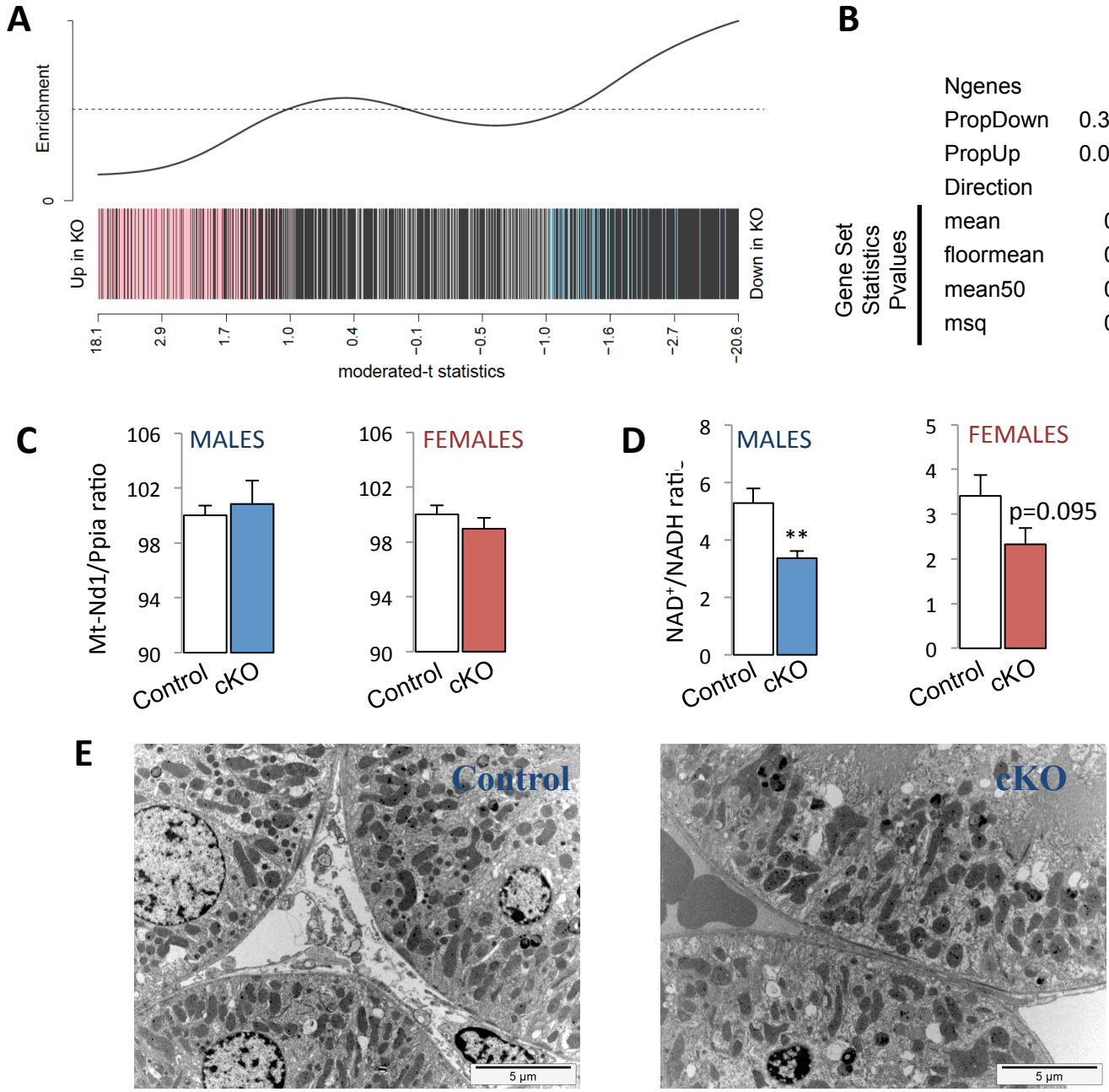
Supplemental Figure 2. Phosphaturia in female and transient phosphaturia in male cKO mice. The excretion rate of Pi was measured in Control and cKO male (top panel) or female (lower panel) mice. Three independent sets of mice were investigated: In the **first set** the urine was collected on the day preceding the 2-weeks period of DOX treatment (Baseline), on days 7 and 14 of DOX treatment (Days DOX) and, on days 7, 14, 21 and 28 following DOX withdrawal (Days post DOX). In this set of mice, a significant phosphaturia was present in female but not in male cKO mice (2-way ANOVA applied to Days DOX and Days post DOX samples). In the **second set** the urine was collected on the day preceding the 2-weeks period of DOX treatment (Baseline) and on days 1 to 7 of DOX treatment (Days DOX, 1st week). In this set of mice there was no difference in the excretion rate of Pi between Control and cKO mice of both sexes (2-way ANOVA). In the **third set** the urine was collected on the day preceding the 2-weeks period of DOX treatment (Baseline) and on days 8 to 14 of DOX treatment (Days DOX, 2nd week). In this set of mice, a significant phosphaturia was present in cKO mice of both sexes (2-way ANOVA). Data are mean±SEM. Collectively these data show that XPR1 deficiency results in phosphaturia in female and transient phosphaturia (during the second week of DOX treatment) in male cKO mice.



Supplemental Figure 3. Magnesuria (**A**) and calciuria (**B**) associated with XPR1 deficiency. The excretion rate of magnesium and calcium were measured in Control and cKO male (left panel) or female (right panel) mice. The 24-hour urines were collected on the day preceding the 2-weeks period of DOX treatment (Baseline), on days 7 and 14 of DOX treatment (Days DOX) and, on days 7, 14, 21 and 28 following DOX withdrawal (Days post DOX). White bars indicate magnesium or calcium excretion rates in Control mice (n=6 for males and n=4 for females). Blue and red bars indicate magnesium or calcium excretion rates in male and female cKO mice, respectively (n=6 for males and n=6 for females). Data are means±SEM. * - indicates statistical significance between Control and cKO mice (*p<0.05; **p<0.01; unpaired t-test). † - indicates statistical significance between excretion rates measured on the day preceding the 2-weeks period of DOX treatment and excretion rates measured during DOX treatment or after the end of DOX treatment (†p<0.05; ††p<0.01; paired t-test).



Supplemental Figure 4. Low urinary pH and polyuria associated with XPR1 deficiency. Urine pH (**A**), urine volume (**B**) and urine osmolality (**C**) were measured in Control and cKO male (left panel) or female (right panel) mice. Urine volume was normalized per gram of body weight. The 24-hour urines were collected four days preceding the 2-weeks period of DOX treatment (Baseline), on days 6, 7, 13 and 14 of DOX treatment (Days DOX) and, on days 6, 7, 14, 20, 21,27 and 28 following DOX withdrawal (Days post DOX). White circles/bars indicate urine pH/volume/osmolality in Control mice (n=6 for males and n=4 for females). Blue and red circles/bars indicate urine pH/volume/osmolality in male and female cKO mice, respectively (n=6 for males and n=6 for females). Data are means±SEM. * - indicates statistical significance between Control and cKO mice (*p<0.05; **p<0.01; ***p<0.001 unpaired t-test). † - indicates statistical significance between the mean of 24-hour urine volumes collected during the baseline period and urine volumes measured during DOX treatment or after the end of DOX treatment (†p<0.05, ††p<0.01; †††p<0.001; paired t-test).



Supplemental Figure 5. Mitochondrial function in Control and cKO mice. **A** and **B:** enrichment of genes coding for mitochondrial proteins among genes down-regulated in kidneys of cKO mice. **(A)** Moderated t-statistics are ranked left to right from largest to smallest. The positions of genes coding for mitochondrial proteins are marked by vertical bars. An enrichment worm shows the relative enrichment of the vertical bars in each part of the plot. **(B)** Enrichment was tested using 'mroast' (Wu et al., Bioinformatics 26, 2176–2182, 2010). The gene set statistics "mean", "floormean", "mean50" and "msq" have different sensitivities to small proportion of the gene set being differentially expressed. With "mean", the set will be statistically significantly different only when the majority of the genes are differentially expressed. The other statistics are sensitive to smaller proportions of differentially expressed genes, if the effects are reasonably large. **C.** The cKO mice exhibit normal mitochondrial biogenesis as shown by quantification of mitochondrial (mtDNA) and nuclear DNA in kidneys of Control and cKO mice. Relative amounts of mtDNA and nuclear DNA were quantitated by qPCR of mitochondrially encoded NADH dehydrogenase 1 (mt-Nd1) and nuclear Ppia (cyclophilin) genes. Data are mean±SEM; n=6; unpaired *t*-test. **D.** The NAD^+-to-NADH ratio, which reflects the oxida-tive phosphorylation-to-glycolysis ratio and/or the status of mitochondrial function, is significantly reduced in kidneys of male cKO mice. Data are mean±SEM. n=9; unpaired *t*-test. **E.** Electron microscopy did not reveal any gross changes in mitochondrial morphology, number or distribution in the proximal tubules of cKO mice.

Supplemental Table 1. Genes differentially expressed in kidneys of Control and cKO mice.
Arbitrary fold-change cut-off of >2 and significance p-values <0.05 were applied for gene selection.
The complete data set is publicly available at GEO through accession number XXX.

Genes upregulated in kidneys of cKO mice.

Gene_Name	AveExp_WT	AveExp_KO	Fold_Change	adj.P.Val
Spaca7	2.953	10.79	228.42	0.00139
D630011A20Rik	2.802	10.46	201.86	0.00062
BC018473	1.296	8.79	180.37	0.01469
Sult4a1	2.845	10.03	145.44	0.00021
A630095E13Rik	0.990	7.68	103.39	0.00052
Vmn1r32	2.449	8.89	87.02	0.00128
Acta1	2.179	8.45	77.32	0.00400
Ano3	2.648	8.74	68.03	0.00284
Krt20	4.871	10.88	64.45	0.00975
Gm37800	0.594	6.57	62.91	0.00031
Casp14	2.312	8.27	62.33	0.00073
Il1f6	1.175	6.98	55.86	0.00722
Havcr1	7.778	13.57	55.39	0.00267
9530053A07Rik	1.571	7.19	49.00	0.00188
Psrc1	5.288	10.82	46.32	0.00051
Defb36	0.990	6.49	45.12	0.00015
Ifi27l2b	1.967	7.44	44.41	0.00031
S100a3	1.387	6.85	43.97	0.00255
Gm7893	2.197	7.55	40.96	0.00422
C4bp	3.840	9.13	39.20	0.00062
Ugt2b35	2.179	7.35	36.03	0.00084
Krt12	0.594	5.72	34.86	0.00045

Genes downregulated in kidneys of cKO mice.

Gene_Name	AveExp_WT	AveExp_KO	Fold_Change	adj.P.Val
Cyp2d12	12.444	4.23	-296.75	0.00015
Gm28438	8.220	1.37	-115.30	0.00031
Slc16a14	11.675	5.08	-96.57	0.00076
Dpys	9.777	3.45	-80.23	0.00224
Slc22a8	14.121	8.08	-65.81	0.00044
Fcamr	10.747	4.71	-65.60	0.00021
Synpr	8.293	2.30	-63.87	0.00295
Gm15563	7.416	1.68	-53.41	0.00097
Gm15883	10.913	5.40	-45.60	0.00015
Cntnap5a	8.247	2.74	-45.56	0.00173
Cyp4a12a	12.877	7.38	-45.22	0.00048
Cyp2e1	17.303	11.90	-42.21	0.00015
Hrg	9.011	3.61	-42.12	0.00053
Pde6a	10.719	5.32	-42.05	0.00031
Nepn	10.963	5.59	-41.42	0.00021
Gm906	6.673	1.49	-36.27	0.00052
Col19a1	7.825	2.65	-36.03	0.00137
Gm4208	9.854	4.69	-35.76	0.00057
Nwd2	5.687	0.79	-29.79	0.00021
Clec2h	13.447	8.68	-27.14	0.00039
F5	9.585	4.92	-25.42	0.00146
Slc34a3	11.818	7.15	-25.37	0.00120

Flnc	5.410	10.46	33.15	0.02048	Gm4477	8.222	3.56	-25.36	0.00010
RP23-421B1.4	2.273	7.29	32.33	0.00229	Fam151a	12.757	8.12	-24.88	0.00046
Serpinb9b	1.967	6.94	31.51	0.00356	Henmt1	7.217	2.58	-24.86	0.00497
Cdkn1a	8.876	13.84	31.10	0.00044	Sectm1b	12.583	7.95	-24.77	0.00015
Mybpc2	3.462	8.38	30.21	0.00077	Ccl28	13.174	8.57	-24.26	0.00010
Il23r	2.394	7.25	29.04	0.00116	Cldn18	6.743	2.16	-23.90	0.00264
Ptprt	2.089	6.94	28.80	0.00929	Gm13412	6.445	1.95	-22.52	0.00151
RP23-337M7.12	0.990	5.82	28.37	0.00293	Ctxn3	10.660	6.17	-22.47	0.01004
Eda2r	7.022	11.78	27.10	0.00014	9030619P08Rik	10.484	6.00	-22.38	0.00192
Dsg1a	0.594	5.30	26.11	0.00095	Trdn	8.245	3.77	-22.30	0.00021
Mir6358	0.594	5.29	25.88	0.00015	G6pc	15.137	10.68	-22.02	0.00015
Lgi2	4.226	8.91	25.68	0.00400	Dnase1	16.576	12.20	-20.76	0.00014
Mmp7	2.179	6.83	25.13	0.00496	Gm22748	7.217	2.87	-20.42	0.00031
Fam212b	5.582	10.20	24.60	0.00046	Gm16004	4.707	0.39	-19.87	0.00142
Fosl1	1.692	6.27	23.91	0.01565	Gm6300	13.085	8.78	-19.80	0.00098
Tmem145	0.990	5.56	23.79	0.00066	Slc13a4	8.755	4.45	-19.71	0.00130
Ugt1a10	5.860	10.39	23.11	0.00039	Slc39a5	10.235	5.97	-19.28	0.00031
Raet1e	2.669	7.16	22.46	0.00062	RP23-227D7.7	8.363	4.10	-19.21	0.00150
Gm5549	1.967	6.43	22.06	0.00082	Srd5a2	9.477	5.25	-18.69	0.00015
Dsg1b	1.175	5.63	21.98	0.00150	Mogat2	10.239	6.22	-16.18	0.00045
RP24-92L16.5	1.571	6.02	21.86	0.00257	Ugt2b38	16.360	12.35	-16.15	0.00010
Padi4	0.990	5.41	21.44	0.00210	RP23-227D7.8	5.741	1.77	-15.71	0.02594
Lcn2	7.307	11.69	20.85	0.00128	Azgp1	13.748	9.79	-15.54	0.00052
Sftpd	0.990	5.35	20.59	0.00204	Ces1g	10.150	6.23	-15.11	0.00287
Phlda3	8.126	12.49	20.56	0.00015	Alb	9.130	5.22	-15.03	0.00173
Slc6a19os	2.805	7.16	20.44	0.01505	Cml1	14.722	10.87	-14.48	0.00052
Lyz1	2.923	7.27	20.30	0.01607	Pnliprp1	6.630	2.78	-14.44	0.03191
Ccdc178	1.783	6.12	20.25	0.00045	Slitrk1	5.857	2.05	-13.98	0.00221
Ccdc60	1.755	6.09	20.14	0.00257	Anxa13	9.181	5.44	-13.40	0.00048

Rasl10b	4.325	8.64	19.95	0.00064	Inmt	16.594	12.85	-13.36	0.00031
Gdap1	1.387	5.66	19.29	0.00122	Gm20713	5.677	1.95	-13.23	0.00289
Gm8104	1.967	6.23	19.19	0.00236	Cyp2f2	7.612	3.92	-12.90	0.00010
Mmp10	0.990	5.22	18.74	0.00243	Gm29012	8.845	5.18	-12.70	0.00031
Cdsn	2.698	6.93	18.73	0.02871	Clip4	10.709	7.05	-12.65	0.00120
Gm12603	0.990	5.19	18.37	0.00220	Cox6a2	7.209	3.58	-12.36	0.00255
Gm1673	2.892	7.07	18.11	0.00724	Gm10384	6.341	2.75	-12.02	0.02072
A230065H16Rik	1.387	5.52	17.57	0.00257	C330002G04Rik	8.281	4.69	-12.01	0.00232
Ms4a2	5.237	9.36	17.43	0.00039	Cyp2d26	12.742	9.16	-11.99	0.00057
Gm13151	1.571	5.67	17.12	0.00264	Gldc	11.568	8.00	-11.89	0.00284
Ntm	4.531	8.56	16.33	0.00031	Gm15354	6.677	3.11	-11.85	0.02355
Timp1	4.308	8.31	16.00	0.00331	Slco1a1	16.201	12.65	-11.76	0.00031
A730017C20Rik	3.852	7.82	15.62	0.00094	Ttr	14.268	10.73	-11.61	0.00205
Mucl1	0.594	4.55	15.50	0.00841	Col6a6	7.573	4.05	-11.53	0.01083
Olfr1314	0.594	4.50	15.01	0.00465	Slc5a12	14.502	11.01	-11.29	0.00230
Gsta1	5.178	9.08	14.99	0.00293	Hpd	12.254	8.77	-11.21	0.00166
Samd5	4.933	8.82	14.80	0.00021	Gabrb3	10.496	7.01	-11.18	0.00137
Sprr2g	2.363	6.25	14.78	0.00443	Cyp4b1	17.790	14.32	-11.06	0.00015
Gm11735	0.594	4.48	14.77	0.03692	Cyp24a1	12.688	9.23	-11.00	0.00630
Serpina10	5.989	9.84	14.45	0.00206	1700119H24Rik	6.799	3.36	-10.88	0.00082
BC107364	1.877	5.71	14.30	0.00598	4930579F01Rik	5.154	1.72	-10.84	0.01648
Foxj1	3.434	7.26	14.20	0.00114	Gm4952	8.568	5.14	-10.75	0.00015
Gjb4	4.260	8.09	14.19	0.00265	Pck1	16.443	13.03	-10.65	0.00118
Cxcl2	2.989	6.80	14.06	0.00233	4930502E18Rik	8.701	5.33	-10.35	0.00132
Tnfrsf9	3.016	6.83	14.03	0.00532	Prodh2	11.313	7.94	-10.33	0.00069
Gm7233	2.151	5.94	13.85	0.00157	AW822252	9.967	6.63	-10.13	0.00163
Mtfr2	4.160	7.95	13.84	0.00264	Angptl7	9.607	6.27	-10.10	0.00053
Akr1c20	5.473	9.24	13.63	0.00039	Gbp10	6.894	3.57	-10.03	0.01858
Tnfrsf12a	10.137	13.90	13.62	0.00073	Slc22a22	14.101	10.78	-10.01	0.00016

Slc13a2os	2.790	6.56	13.60	0.03252	Ighv1-15	5.247	1.96	-9.75	0.02606
Aldh1a1	9.779	13.54	13.58	0.00015	Ces2h	8.010	4.74	-9.64	0.00295
Atf3	6.833	10.59	13.55	0.00094	C4a	8.057	4.79	-9.64	0.00492
Gm21807	2.394	6.15	13.54	0.00317	Lipo2	8.376	5.13	-9.51	0.00094
Tacr3	2.179	5.93	13.44	0.00485	Ces1e	13.104	9.86	-9.51	0.00017
Npy2r	2.089	5.83	13.41	0.01435	RP23-381C14.2	6.576	3.35	-9.34	0.00098
Trem2	3.625	7.35	13.26	0.00148	Acmsd	13.463	10.25	-9.27	0.00113
Ddias	6.591	10.31	13.19	0.00015	Gm5424	9.817	6.63	-9.10	0.00056
Gtse1	5.339	9.01	12.74	0.00215	Tm4sf4	7.349	4.18	-9.02	0.01291
Ankrd1	7.196	10.86	12.66	0.00139	Chrna4	10.746	7.58	-8.99	0.02470
St6galnac1	0.594	4.26	12.65	0.03813	Odc1	16.948	13.79	-8.90	0.00032
Nlrp1a	3.691	7.35	12.64	0.00048	Astn1	6.738	3.61	-8.76	0.00266
Asb5	5.846	9.48	12.39	0.00335	Car14	12.729	9.60	-8.75	0.00062
Lif	4.734	8.35	12.24	0.00455	Etnppl	10.871	7.74	-8.74	0.00015
Smpdl3b	6.350	9.96	12.17	0.00216	Nap1l5	8.825	5.71	-8.69	0.00016
Rem2	3.277	6.88	12.12	0.00053	Hapln1	7.607	4.49	-8.67	0.00283
Fcrls	4.023	7.59	11.84	0.00518	Gm7278	6.986	3.87	-8.65	0.00162
Clvs2	2.394	5.96	11.83	0.00603	Gm128	7.763	4.65	-8.64	0.00028
S100a6	10.814	14.37	11.79	0.00598	Gm9115	7.472	4.39	-8.49	0.00053
Hist1h1d	1.998	5.55	11.73	0.01685	Gm13936	5.831	2.75	-8.44	0.01752
Ccl11	4.637	8.17	11.59	0.00106	Stab2	8.913	5.88	-8.18	0.00126
Klrg2	6.583	10.11	11.53	0.00010	Corin	8.236	5.21	-8.17	0.00610
Il1rn	4.310	7.81	11.34	0.01316	Sox2ot	7.557	4.53	-8.16	0.00251
Ttn	4.420	7.91	11.23	0.04893	Serpinf2	14.598	11.60	-8.01	0.00031
Gml	2.151	5.63	11.14	0.01028	Ankrd34b	6.964	3.97	-7.99	0.00092
1700016K19Rik	1.783	5.25	11.05	0.00210	Trim63	10.727	7.73	-7.96	0.00284
1700007K13Rik	5.258	8.71	10.94	0.00063	4930533I22Rik	8.027	5.04	-7.91	0.01316
Chil4	4.087	7.54	10.93	0.00586	Gm11788	6.541	3.56	-7.88	0.01926
Serpina7	3.824	7.27	10.89	0.00169	RP23-57A17.2	7.067	4.09	-7.87	0.00166

Bst1	10.167	13.60	10.80	0.00065	Nr1i3	5.127	2.16	-7.80	0.04190
Gm13398	4.106	7.54	10.79	0.00060	Cckar	13.358	10.40	-7.77	0.00024
Tmem202	2.732	6.15	10.70	0.00669	Keg1	17.485	14.54	-7.70	0.00056
Gm11682	0.990	4.40	10.64	0.02083	Cyp2j13	14.840	11.90	-7.65	0.00056
Gm13067	2.363	5.77	10.57	0.00805	RP23-287M3.2	7.443	4.51	-7.65	0.02101
Ntf5	4.737	8.13	10.47	0.00097	Sugct	13.249	10.33	-7.54	0.00016
Col11a2	5.160	8.54	10.40	0.00227	Ak4	14.923	12.01	-7.54	0.00356
Nkd2	6.672	10.03	10.22	0.00053	Rnf24	12.925	10.03	-7.46	0.04944
Retnla	1.387	4.73	10.17	0.01861	Gm13528	7.354	4.47	-7.36	0.00629
Grem1	3.984	7.33	10.17	0.00317	Xpr1	12.790	9.92	-7.30	0.00410
Zfp365	4.114	7.46	10.15	0.00138	Snx31	11.664	8.81	-7.24	0.00057
Sh2d5	1.916	5.26	10.14	0.01881	Cndp1	12.522	9.68	-7.19	0.00133
A930001A20Rik	3.858	7.19	10.07	0.00106	Slc34a1	17.744	14.90	-7.19	0.00552
Avil	4.511	7.82	9.94	0.00197	Slc51a	12.267	9.44	-7.08	0.00128
Tpx2	6.175	9.48	9.89	0.00053	Ido2	9.545	6.72	-7.07	0.00371
S100z	2.312	5.61	9.86	0.02732	Gm38204	6.311	3.50	-7.04	0.00603
Tmprss7	2.548	5.83	9.75	0.00426	Dhtkd1	11.519	8.71	-7.02	0.00662
Prtn3	1.967	5.24	9.67	0.00685	Cml5	11.713	8.91	-6.98	0.00259
Tssk6	3.250	6.49	9.45	0.00218	Ces1f	16.157	13.36	-6.97	0.00070
Celf5	2.548	5.74	9.15	0.01411	Fmo1	15.640	12.84	-6.95	0.00020
Clcf1	7.912	11.09	9.05	0.00277	Aspdh	12.662	9.87	-6.93	0.00031
Guca1a	4.317	7.49	9.05	0.01367	Agxt2	12.815	10.02	-6.93	0.00015
Popdc3	5.145	8.31	9.00	0.00873	Slc18a1	14.186	11.40	-6.91	0.00200
Medag	2.975	6.14	9.00	0.02351	Afmid	11.011	8.24	-6.83	0.00015
Calcb	1.967	5.13	8.96	0.01362	Ass1	16.303	13.53	-6.82	0.00082
Lypd3	2.336	5.50	8.95	0.01004	Ksr2	9.261	6.50	-6.78	0.04572
Xirp2	2.151	5.30	8.88	0.01201	Gm10522	7.666	4.91	-6.77	0.00021
Ngf	7.461	10.57	8.63	0.00183	Gm12962	7.239	4.50	-6.70	0.00929
Gstm6	6.761	9.87	8.62	0.00072	Nxpe3	11.134	8.39	-6.68	0.00174

Apod	4.924	8.03	8.60	0.01960	Ccdc150	6.357	3.62	-6.68	0.00345
Gm16174	3.340	6.43	8.53	0.02852	Pzp	13.298	10.57	-6.60	0.01102
Prss35	6.091	9.18	8.50	0.00664	Cyp4a14	9.566	6.84	-6.59	0.01552
Ndn	6.899	9.98	8.48	0.00120	Gm16755	7.407	4.69	-6.59	0.00692
Meis3	6.873	9.96	8.48	0.00063	Apcs	6.516	3.81	-6.52	0.00230
Lpo	4.265	7.34	8.45	0.01828	Tmem26	10.088	7.39	-6.49	0.00117
Gdf15	8.734	11.81	8.43	0.00121	Lin7a	8.479	5.78	-6.47	0.00089
Htr3a	5.204	8.28	8.42	0.00039	Prok1	10.981	8.29	-6.47	0.00136
Fam131c	7.407	10.48	8.39	0.00039	Kmo	13.723	11.03	-6.45	0.00021
Vwa7	4.845	7.91	8.37	0.00117	Prodh	13.528	10.86	-6.37	0.00120
Gm15867	2.732	5.77	8.24	0.02372	0610031O16Rik	9.935	7.26	-6.36	0.00073
9130230L23Rik	4.829	7.87	8.22	0.00196	Slc22a6	15.765	13.10	-6.34	0.00433
Plekhs1	3.413	6.45	8.18	0.00196	Acsm2	18.561	15.90	-6.32	0.00130
Lgals3	10.978	14.00	8.11	0.00592	Cml3	9.493	6.83	-6.32	0.00062
Ghrhr	4.136	7.14	8.02	0.00216	Ccdc170	7.182	4.52	-6.31	0.00484
Clec2l	3.385	6.39	8.02	0.00538	Hmcn2	9.544	6.89	-6.29	0.00733
Slc7a12	7.661	10.66	8.01	0.02052	Glt1d1	10.456	7.81	-6.28	0.00983
Sct	2.732	5.73	8.00	0.01465	Gm11837	8.741	6.12	-6.14	0.00992
Gm26559	3.138	6.13	7.98	0.00400	Kcnj12	6.510	3.89	-6.14	0.01026
Cdkn2b	7.338	10.32	7.90	0.00173	Bbox1	7.263	4.68	-5.99	0.00189
Snca	7.989	10.96	7.82	0.00047	Pecr	15.166	12.58	-5.99	0.00056
Ttc16	4.616	7.58	7.81	0.00521	Tmem169	9.935	7.36	-5.95	0.00064
Lyz2	10.956	13.88	7.61	0.01766	Unc5c	8.981	6.43	-5.86	0.01554
1500015O10Rik	5.583	8.51	7.59	0.01386	Bhmt	7.109	4.56	-5.83	0.00062
2310014F06Rik	4.153	7.06	7.51	0.00210	Slc22a30	13.993	11.45	-5.82	0.00082
Tbpl2	2.548	5.42	7.32	0.04761	Tmprss9	7.294	4.75	-5.82	0.00332
Dynlrb2	3.837	6.69	7.23	0.00204	Pyroxd2	13.114	10.57	-5.81	0.00255
Igfbp2	7.232	10.08	7.21	0.00381	4933421A08Rik	5.983	3.45	-5.81	0.02429
Gm9732	3.350	6.19	7.14	0.04555	Notum	10.180	7.65	-5.78	0.00095

Spaca1	2.363	5.20	7.13	0.04465	Tgm1	10.819	8.30	-5.74	0.00720
Scx	6.369	9.18	7.04	0.00680	Col27a1	11.463	8.94	-5.74	0.03732
Dusp8	6.480	9.29	7.03	0.01226	Ugt1a7c	14.070	11.56	-5.71	0.00236
Enkur	4.753	7.55	6.96	0.00070	Dleu7	10.758	8.24	-5.71	0.00072
Plaur	8.372	11.17	6.95	0.00054	Slc13a1	14.493	11.99	-5.67	0.00039
Ucp3	4.662	7.46	6.95	0.00376	Gm38319	7.559	5.06	-5.66	0.02186
Akr1b8	8.158	10.95	6.91	0.00100	Ces2g	8.759	6.26	-5.65	0.00147
Masp1	5.552	8.33	6.88	0.00431	Hykk	14.879	12.39	-5.61	0.00665
Trim31	2.557	5.34	6.87	0.04247	Acy3	17.125	14.65	-5.56	0.00062
Prss16	3.778	6.56	6.86	0.00216	Bnc2	10.388	7.92	-5.55	0.00685
Ercc6l	5.062	7.83	6.83	0.00057	Adra2b	13.160	10.70	-5.50	0.00089
Pabpc1l	3.371	6.14	6.81	0.00812	Bco2	11.047	8.59	-5.48	0.00034
Snph	2.273	5.03	6.77	0.04132	Aacs	13.103	10.65	-5.47	0.00809
Tnfsf8	3.128	5.88	6.75	0.00063	Alpl	13.248	10.81	-5.44	0.00429
Hspb1	8.320	11.07	6.72	0.00081	Bcl11b	8.731	6.30	-5.39	0.02916
Wbscr17	3.382	6.13	6.71	0.02000	Fam163a	9.424	7.00	-5.38	0.00214
2310043M15Rik	3.716	6.46	6.71	0.00287	Thnsl2	14.179	11.75	-5.37	0.00056
Rprm	4.374	7.11	6.65	0.01170	Amacr	14.930	12.51	-5.37	0.00031
Jazf1	5.504	8.23	6.62	0.00045	Aoah	12.295	9.88	-5.35	0.00056
Serp2	4.917	7.63	6.57	0.02088	Pik3c2g	9.362	6.94	-5.35	0.00053
Anxa3	9.973	12.68	6.54	0.00034	Pigr	13.718	11.31	-5.31	0.00129
Ccr1	5.510	8.22	6.54	0.00192	Errfi1	15.333	12.95	-5.22	0.00145
Serpina3n	4.772	7.46	6.45	0.00157	Cyp2d9	14.053	11.67	-5.20	0.00145
Gadd45b	7.482	10.17	6.45	0.00225	1110032F04Rik	6.804	4.43	-5.18	0.00132
Tmem255b	3.674	6.35	6.39	0.00198	Mylk3	8.294	5.92	-5.17	0.00216
Spink12	2.151	4.82	6.34	0.02916	St8sia1	13.567	11.20	-5.15	0.00251
Gm13056	4.238	6.90	6.32	0.04247	Sucnr1	12.526	10.16	-5.15	0.00090
Fndc4	7.781	10.44	6.32	0.00413	Slc16a2	13.664	11.30	-5.13	0.00629
5730437C11Rik	4.501	7.15	6.28	0.01316	Slc26a1	13.204	10.85	-5.11	0.00293

BC046401	3.361	5.98	6.16	0.00583	Pbld2	14.443	12.10	-5.07	0.00060
Clca3a2	5.090	7.71	6.14	0.00171	Macrod2	14.747	12.42	-5.02	0.00121
1700016C15Rik	11.287	13.90	6.12	0.02136	Gjb2	13.059	10.73	-5.02	0.00063
Fcrl1	3.941	6.54	6.08	0.00410	Ces2c	13.627	11.31	-5.00	0.00259
Defb26	3.555	6.16	6.07	0.00159	Gm11128	12.618	10.30	-4.99	0.00152
2200002D01Rik	8.754	11.35	6.06	0.02001	Gm37824	10.039	7.72	-4.98	0.01706
Ms4a6d	5.018	7.61	6.05	0.01766	1700067K01Rik	6.707	4.40	-4.93	0.00547
Mcmdc2	4.438	7.03	6.04	0.00092	Slc29a3	12.095	9.79	-4.93	0.02911
Hist1h4h	7.399	9.99	6.03	0.02995	Pah	16.502	14.21	-4.91	0.00057
Hspb8	8.656	11.24	6.00	0.00274	Cyp2a5	13.366	11.08	-4.88	0.00695
Fam171b	4.453	7.03	5.95	0.01054	Snx29	11.835	9.55	-4.88	0.01083
Ankef1	2.953	5.52	5.91	0.01308	Gm11827	8.291	6.01	-4.86	0.00129
Tslp	4.153	6.72	5.91	0.00098	Gm15990	6.367	4.10	-4.82	0.02593
Il5ra	9.360	11.90	5.81	0.00139	Slc28a1	9.982	7.72	-4.80	0.01712
Creb5	5.852	8.38	5.78	0.01169	Lpl	16.227	13.96	-4.80	0.00064
Capsl	4.979	7.50	5.74	0.00606	Gm15879	9.840	7.59	-4.77	0.00090
Gprc5a	7.673	10.19	5.73	0.00070	Stxbp5l	9.561	7.31	-4.76	0.01739
Acot5	2.853	5.36	5.70	0.00737	Tfpi2	10.052	7.80	-4.76	0.00685
Ptrh1	7.278	9.78	5.67	0.00746	Pdp2	11.458	9.23	-4.68	0.00392
Slc25a24	8.672	11.15	5.55	0.00062	B3galt5	11.841	9.62	-4.66	0.02145
Ripk3	5.988	8.44	5.49	0.00163	Slc13a2	11.748	9.53	-4.66	0.00508
Gulo	3.534	5.99	5.49	0.01155	Gm37844	7.379	5.17	-4.64	0.01291
4930427A07Rik	4.751	7.20	5.47	0.02101	Adh1	15.113	12.90	-4.63	0.00251
Golm1	9.397	11.85	5.46	0.00171	Defb19	5.585	3.37	-4.63	0.02427
Ltbp2	3.990	6.43	5.42	0.00446	Igfbp1	9.014	6.80	-4.62	0.00548
Muc19	2.708	5.14	5.40	0.03084	Pank1	14.732	12.52	-4.62	0.00257
Bcl2a1b	4.093	6.53	5.40	0.03359	Ankrd65	8.545	6.34	-4.60	0.00146
Gm11613	6.197	8.62	5.36	0.00214	9630013D21Rik	9.763	7.56	-4.60	0.00606
Ear2	3.961	6.38	5.36	0.02636	Igfbp4	15.067	12.87	-4.59	0.00210

Chrna2	4.065	6.49	5.35	0.01170	Slc7a8	12.139	9.94	-4.59	0.04557
Dio2	6.023	8.44	5.34	0.02036	Fbxo40	11.781	9.58	-4.58	0.00467
Ephx1	11.644	14.05	5.30	0.00284	Rhobtb1	11.324	9.13	-4.58	0.00606
Steap1	8.112	10.51	5.28	0.00158	Slc25a25	12.836	10.64	-4.57	0.01444
1600010M07Rik	3.096	5.50	5.28	0.01877	Degs2	12.152	9.97	-4.55	0.00611
Adgra1	3.458	5.84	5.23	0.02541	Apom	12.177	10.00	-4.52	0.00121
Olfr920	4.420	6.81	5.23	0.02685	Slc17a3	15.789	13.61	-4.52	0.00158
Chrnb1	7.784	10.17	5.22	0.00147	Akr1c14	15.351	13.18	-4.50	0.00142
Abcb4	4.224	6.61	5.22	0.00484	Osbp2	10.773	8.61	-4.49	0.01121
Sult2b1	5.543	7.92	5.21	0.00812	D630029K05Rik	14.063	11.90	-4.49	0.00062
Bco1	7.575	9.95	5.21	0.00142	Als2cr12	8.649	6.48	-4.49	0.00229
2610020C07Rik	4.081	6.46	5.20	0.00081	Epb4.1l3	10.905	8.74	-4.48	0.01825
Spag6	6.003	8.38	5.20	0.00046	Fgb	5.954	3.79	-4.47	0.01697
Gm16201	3.250	5.62	5.18	0.00873	Slc22a28	13.624	11.46	-4.47	0.00153
Fbxo2	6.125	8.50	5.18	0.00257	Fut9	15.742	13.58	-4.47	0.02551
Brca1	5.513	7.88	5.17	0.01666	Gatm	16.022	13.88	-4.43	0.00174
Cpt1c	6.678	9.04	5.15	0.00150	Adhfe1	14.887	12.74	-4.42	0.00031
Upk3bl	3.443	5.80	5.12	0.01600	Acsm3	16.817	14.69	-4.38	0.00197
Msr1	5.431	7.78	5.11	0.00905	Acnat2	11.893	9.76	-4.37	0.01260
Ckap2	6.269	8.62	5.10	0.00150	Pcx	14.432	12.31	-4.36	0.01159
Gzmm	5.019	7.37	5.09	0.00675	Gm16348	6.444	4.32	-4.36	0.00340
Capg	10.052	12.39	5.07	0.00997	Aldh3b3	7.705	5.58	-4.35	0.00174
Myof	9.848	12.19	5.07	0.01882	Chst11	12.201	10.09	-4.32	0.01752
Gm15471	5.713	8.04	5.03	0.01348	Gm11127	9.136	7.03	-4.32	0.02182
Fgf7	4.665	6.99	5.01	0.00153	Slc46a3	11.200	9.09	-4.31	0.00127
Lppr3	5.061	7.38	5.00	0.00603	Acsm1	13.957	11.85	-4.30	0.00116
Gm32200	3.594	5.91	4.98	0.00528	Car9	9.966	7.86	-4.29	0.00330
Kcnu1	4.622	6.92	4.93	0.00202	Galnt11	15.945	13.85	-4.28	0.00130
Akr1c12	8.098	10.40	4.92	0.00207	Eci3	14.594	12.50	-4.28	0.00315

Edn1	6.443	8.74	4.91	0.00387	Slc22a12	17.063	14.97	-4.28	0.00133
Ugt1a2	5.462	7.75	4.89	0.02515	March10	6.710	4.62	-4.27	0.00254
Ccne1	5.418	7.71	4.89	0.00163	Cyp7b1	14.606	12.51	-4.27	0.00166
Nrn1	3.138	5.42	4.88	0.01554	Ppp1r16b	12.180	10.09	-4.24	0.02959
Hist1h2ac	3.492	5.78	4.87	0.00720	BC026585	14.759	12.67	-4.24	0.00048
Gm13418	3.289	5.57	4.87	0.01730	Slc7a9	13.263	11.18	-4.24	0.00058
Ddit4l	9.813	12.09	4.86	0.00566	Syn3	9.276	7.20	-4.23	0.00159
Dio1	10.476	12.76	4.86	0.00401	Pm20d1	12.446	10.37	-4.21	0.00685
Ska3	3.990	6.27	4.86	0.00289	Hnf4g	9.916	7.85	-4.18	0.00192
Mgp	12.236	14.52	4.86	0.01129	Gm12999	8.587	6.53	-4.17	0.00404
Cryab	13.215	15.48	4.80	0.00583	Cryz	15.482	13.42	-4.17	0.00045
Dpysl3	9.505	11.77	4.80	0.00739	Spp2	14.086	12.03	-4.16	0.00698
Camk1g	6.535	8.80	4.79	0.00174	Ppara	11.551	9.50	-4.15	0.01730
Oit1	5.898	8.16	4.79	0.00934	Kcnj15	16.187	14.14	-4.14	0.00485
Gdpd2	3.413	5.67	4.79	0.00255	BC089597	11.976	9.93	-4.13	0.01367
Cdk1	6.924	9.17	4.74	0.01395	Gm12195	6.666	4.62	-4.12	0.01274
Vtcn1	6.431	8.67	4.73	0.00196	Hebp1	12.806	10.77	-4.09	0.00685
Cbr3	6.430	8.67	4.71	0.04962	Dgkg	9.214	7.19	-4.08	0.00284
RP23-400L4.7	5.494	7.73	4.71	0.01711	Nat8	14.234	12.21	-4.07	0.00782
Ccdc13	3.159	5.39	4.70	0.00221	Fmo5	13.827	11.81	-4.05	0.00057
Gm11974	8.130	10.36	4.68	0.01079	Slc25a42	12.570	10.56	-4.04	0.02023
Slc6a12	7.337	9.56	4.67	0.00426	Adamts15	9.767	7.75	-4.04	0.02729
Nnmt	6.580	8.80	4.65	0.02336	Bhmt2	13.251	11.24	-4.03	0.00114
Egr2	4.683	6.90	4.64	0.01037	Gpr137b-ps	10.386	8.38	-4.02	0.00121
Bche	5.491	7.69	4.59	0.00318	Aadat	14.804	12.80	-4.02	0.00284
Bcl2a1d	3.818	6.02	4.59	0.02351	Slc30a2	12.674	10.67	-4.00	0.00174
Spp1	15.624	17.82	4.58	0.00036	Slc16a9	13.808	11.81	-3.99	0.00685
Aldh1a2	7.651	9.85	4.58	0.00126	Gm4131	8.914	6.92	-3.98	0.02128
Tnfrsf10b	7.583	9.78	4.58	0.00093	Kif20b	11.992	10.00	-3.97	0.01972

D230017M19Rik	5.399	7.59	4.58	0.00174	Tbx10	7.953	5.97	-3.96	0.02012
Cd276	4.260	6.45	4.57	0.01379	Ldhd	16.654	14.67	-3.96	0.00230
Clec18a	5.308	7.50	4.56	0.00216	Tmem64	13.359	11.37	-3.96	0.00807
Pbx4	5.155	7.34	4.55	0.01367	Nox4	15.148	13.16	-3.96	0.00361
Sag	3.534	5.72	4.55	0.00177	Slc6a19	14.887	12.91	-3.93	0.00508
Cygb	8.992	11.17	4.54	0.00156	Cyp4a10	13.937	11.96	-3.93	0.00974
Panx1	6.252	8.43	4.53	0.00289	Enpp6	12.829	10.87	-3.90	0.00398
Gxylt2	5.640	7.81	4.49	0.00761	Acsm5	13.386	11.43	-3.89	0.00354
C1qb	10.274	12.44	4.49	0.02251	Gm26513	6.454	4.50	-3.88	0.00607
Svop	4.524	6.69	4.48	0.00293	RP24-471B15.3	8.045	6.09	-3.87	0.03818
Slc17a2	3.975	6.14	4.48	0.00913	Ugt1a6a	7.477	5.53	-3.86	0.00174
Ccl2	5.136	7.29	4.46	0.01715	Mfsd2a	11.293	9.35	-3.84	0.02197
P2ry12	4.607	6.76	4.46	0.01727	Mcm10	9.069	7.13	-3.83	0.00446
Isg20	7.086	9.24	4.46	0.00433	Fhl1	14.305	12.37	-3.83	0.00074
Zkscan2	3.875	6.03	4.46	0.02110	Haao	14.109	12.17	-3.83	0.01143
Apitd1	5.540	7.70	4.46	0.02296	Rab11fip3	15.222	13.29	-3.82	0.00255
Guca2a	6.795	8.95	4.45	0.02516	Aass	15.125	13.19	-3.82	0.00120
5830416P10Rik	6.140	8.29	4.45	0.00274	Dnajc12	14.575	12.64	-3.81	0.02187
Gm13212	4.047	6.19	4.41	0.01609	Gstp2	6.958	5.03	-3.80	0.01219
Socs3	7.147	9.29	4.41	0.00209	Fmo2	15.260	13.33	-3.80	0.00157
Ccdc109b	5.883	8.02	4.41	0.01062	Timd2	12.829	10.90	-3.80	0.00427
1110046J04Rik	5.434	7.56	4.38	0.02331	Upb1	13.911	11.99	-3.79	0.00063
Efnb3	4.163	6.29	4.37	0.01424	Slc13a3	14.677	12.76	-3.79	0.01730
Ttc39a	6.023	8.15	4.37	0.00067	Aldh6a1	16.424	14.51	-3.78	0.00205
Scin	11.652	13.78	4.36	0.00031	4931406C07Rik	16.284	14.37	-3.78	0.00062
Nxnl2	3.858	5.98	4.35	0.01169	3110045C21Rik	10.284	8.37	-3.77	0.01521
Arl11	4.753	6.87	4.33	0.01381	Ces2b	9.902	7.99	-3.76	0.00174
Platr22	4.286	6.40	4.32	0.00102	Phyhd1	7.902	5.99	-3.76	0.01560
Them6	9.575	11.69	4.32	0.00693	Sypl2	11.650	9.74	-3.76	0.04215

Efcab11	3.879	5.98	4.30	0.03750	Gm30097	6.276	4.37	-3.75	0.00584
Gm9530	4.214	6.32	4.29	0.00153	Aspg	11.988	10.08	-3.75	0.00280
Dlgap5	4.522	6.62	4.28	0.00241	Gm11766	8.367	6.46	-3.74	0.01274
Itih3	6.538	8.63	4.28	0.01975	Slc22a26	11.250	9.35	-3.74	0.00473
Kdelr3	7.418	9.51	4.26	0.01571	Slc6a20b	14.137	12.24	-3.74	0.00181
Slc2a6	4.511	6.60	4.25	0.00586	Hnf4aos	9.671	7.77	-3.73	0.01328
Adamtsl5	6.575	8.66	4.25	0.00413	Tc2n	10.713	8.83	-3.69	0.00929
C6	4.610	6.69	4.22	0.00918	Ceacam1	10.053	8.17	-3.69	0.00963
Wnt10a	3.890	5.97	4.22	0.00545	Igfals	8.984	7.10	-3.69	0.00250
Lrrc26	3.473	5.54	4.19	0.01419	Rdh16	13.417	11.53	-3.69	0.00056
Pcsk9	6.725	8.79	4.18	0.01874	Abhd3	13.234	11.36	-3.67	0.00201
C1qa	9.992	12.06	4.18	0.01456	Cd36	16.077	14.20	-3.66	0.00097
Rap1gapos	3.368	5.43	4.17	0.04388	Car12	13.893	12.03	-3.65	0.00858
Klc3	7.166	9.22	4.16	0.01034	Ncam2	7.151	5.29	-3.64	0.01988
Tmem43	10.047	12.10	4.16	0.00216	C8a	11.485	9.62	-3.64	0.01388
H2-DMa	9.063	11.11	4.14	0.01136	Acox1	16.149	14.29	-3.64	0.00709
Gm14161	4.992	7.04	4.14	0.01083	Selenbp2	10.508	8.65	-3.63	0.00228
Adamts4	5.085	7.14	4.14	0.01758	Adgrg4	8.963	7.11	-3.61	0.00142
Tnfaip8l2	6.167	8.21	4.13	0.01766	Serpina1b	10.485	8.63	-3.61	0.00699
Cp	11.718	13.76	4.12	0.00048	Slc25a21	10.469	8.62	-3.60	0.00070
Il18r1	6.353	8.39	4.11	0.00158	Bex4	9.140	7.30	-3.59	0.01600
Lgi4	5.070	7.11	4.10	0.00265	Acss1	13.512	11.68	-3.57	0.01585
RP23-288B16.4	4.150	6.19	4.10	0.03058	Gm16157	6.775	4.95	-3.55	0.00316
Inhbb	7.097	9.13	4.10	0.00196	Gm6999	7.398	5.57	-3.55	0.00171
Pdlim7	8.708	10.73	4.08	0.00125	Hsd17b2	10.732	8.91	-3.55	0.03426
Tlr2	7.489	9.51	4.05	0.00174	Arsg	11.968	10.14	-3.54	0.00517
Rap2b	7.760	9.77	4.04	0.00070	Lrat	8.322	6.50	-3.54	0.02641
Lox	7.919	9.93	4.04	0.01687	Lpar3	10.840	9.02	-3.53	0.00157
Aoc1	7.888	9.90	4.04	0.00088	Slc35f1	8.761	6.94	-3.53	0.00320

Sprr1a	7.878	9.89	4.03	0.00086	Lrp2	16.370	14.55	-3.52	0.02331
Gdpd1	7.062	9.07	4.03	0.00142	1700034P13Rik	7.669	5.86	-3.51	0.03421
Pf4	5.611	7.62	4.02	0.03606	Gys2	10.109	8.30	-3.51	0.00618
H2-Ab1	12.225	14.23	4.01	0.01204	Ctnna2	8.152	6.34	-3.50	0.01121
Ms4a6b	7.452	9.45	4.01	0.00878	Ccdc6	12.966	11.16	-3.50	0.04751
Ccng1	14.743	16.75	4.01	0.00255	Ldlrad3	11.612	9.81	-3.49	0.00905
Icam4	3.187	5.19	4.01	0.03537	Erich4	12.477	10.67	-3.49	0.00467
Fam46b	7.028	9.03	4.00	0.00213	Rnf152	11.799	10.00	-3.48	0.01651
Ube2t	5.075	7.07	3.98	0.02101	Asl	13.789	11.99	-3.47	0.00285
Gpr22	3.788	5.77	3.96	0.01716	Nlrp6	13.006	11.21	-3.47	0.03605
Homer3	8.710	10.69	3.96	0.00095	Slc7a13	16.552	14.76	-3.47	0.00483
1700011H14Rik	13.277	15.26	3.94	0.03426	Cym	7.294	5.50	-3.46	0.01316
Gdpd3	9.153	11.13	3.94	0.01784	Slc22a7	13.880	12.09	-3.45	0.02382
Cyr61	11.051	13.03	3.94	0.02294	Ugt3a2	17.234	15.45	-3.45	0.00463
Vcam1	9.504	11.48	3.93	0.00114	Npy6r	6.517	4.73	-3.44	0.04787
Dusp14	9.962	11.94	3.93	0.00065	Tfrc	12.992	11.21	-3.44	0.01115
Hesx1	5.058	7.03	3.93	0.04735	Tnfaip8	15.129	13.35	-3.43	0.00147
Ier5	9.293	11.27	3.93	0.00056	C1rl	11.314	9.54	-3.43	0.00127
2310007B03Rik	7.051	9.02	3.92	0.00273	RP23-418O21.3	10.219	8.44	-3.42	0.01274
Fbxo24	3.657	5.62	3.90	0.02882	Hlf	12.726	10.95	-3.42	0.02033
Adgrg3	8.066	10.02	3.88	0.00728	Aldh7a1	14.177	12.41	-3.41	0.00401
Gm13657	3.875	5.83	3.88	0.03882	Lama3	10.874	9.11	-3.40	0.03017
Serpine2	10.147	12.10	3.86	0.00467	Slco1a6	14.530	12.76	-3.40	0.00216
Prkag3	8.946	10.89	3.86	0.00639	Nlrc4	9.399	7.64	-3.37	0.00368
Wnt16	4.465	6.41	3.86	0.00492	Gm27184	6.106	4.35	-3.37	0.00215
Dok3	6.770	8.72	3.85	0.00108	Cyp2a4	15.649	13.90	-3.37	0.00368
Mroh3	3.674	5.62	3.85	0.00596	Fads2	12.357	10.61	-3.36	0.01090
Scn1b	8.879	10.82	3.85	0.00133	Slc7a7	13.128	11.38	-3.36	0.00280
Cntn1	6.210	8.14	3.82	0.00132	Gsap	10.894	9.15	-3.36	0.00273

Acsbg1	5.391	7.32	3.82	0.00969	Lipo1	13.255	11.51	-3.35	0.00473
H2-DMb1	9.921	11.85	3.82	0.01381	Mttp	10.467	8.72	-3.35	0.00703
Fcgr3	8.546	10.46	3.76	0.02506	Col8a1	8.820	7.08	-3.35	0.00610
Itgam	6.290	8.20	3.76	0.00138	Tnfsf10	13.161	11.42	-3.34	0.00303
Oasl1	6.768	8.68	3.76	0.00458	Ubiad1	12.742	11.00	-3.34	0.00181
Gm16537	3.990	5.90	3.75	0.04388	Them7	13.499	11.76	-3.33	0.00062
Gm37795	7.177	9.08	3.75	0.03070	RP23-370A2.6	6.554	4.82	-3.32	0.00889
C1qc	9.813	11.71	3.73	0.00898	Hao2	14.125	12.39	-3.32	0.00243
Prune2	5.121	7.02	3.73	0.01119	Fam210a	13.601	11.87	-3.32	0.00289
Htra3	4.898	6.79	3.72	0.00259	Hnf4a	15.601	13.87	-3.32	0.02529
Pmaip1	10.234	12.13	3.71	0.04902	Ces1d	14.732	13.01	-3.31	0.00222
Gpnmb	4.142	6.03	3.70	0.00700	9130204K15Rik	7.015	5.29	-3.31	0.04138
Ssbp4	9.734	11.62	3.70	0.00200	Spock3	6.920	5.20	-3.30	0.01982
Apoa4	9.567	11.45	3.69	0.02020	Sap30	12.093	10.37	-3.29	0.00126
Ms4a6c	7.230	9.11	3.68	0.02916	Hmgcs1	15.285	13.57	-3.29	0.01560
Ncam1	6.770	8.65	3.68	0.00825	B3galnt1	10.324	8.61	-3.29	0.00389
Lrfn3	6.634	8.51	3.67	0.02959	Mfsd7c	10.167	8.45	-3.28	0.00815
Kifc1	5.345	7.22	3.66	0.00573	Gm10804	13.057	11.35	-3.27	0.00213
Tert	6.452	8.32	3.66	0.00063	Snhg11	13.479	11.77	-3.27	0.03824
Tnnt2	4.217	6.09	3.65	0.01108	Rgl1	13.981	12.28	-3.26	0.00807
Ighg2c	7.339	9.20	3.64	0.04787	Acox3	14.192	12.49	-3.25	0.01782
Cebpd	7.475	9.34	3.64	0.02511	Tmc3	6.811	5.11	-3.25	0.04310
Plekho1	9.712	11.58	3.64	0.02246	Agps	15.459	13.76	-3.25	0.00801
Rgs16	4.249	6.11	3.64	0.01381	Slc22a29	8.326	6.63	-3.25	0.00812
Lrrc23	4.813	6.68	3.64	0.01360	Slc19a3	10.472	8.77	-3.25	0.00377
Aurkb	5.223	7.09	3.64	0.00120	Smco3	7.416	5.72	-3.25	0.00575
Cd14	8.157	10.02	3.63	0.01766	Rpp40	10.420	8.72	-3.25	0.01982
4930461G14Rik	8.614	10.47	3.63	0.04732	Acot12	14.252	12.56	-3.24	0.00217
Ifi205	4.961	6.82	3.62	0.03496	Akr1c18	10.757	9.06	-3.24	0.03070

Nefm	3.939	5.79	3.61	0.01388	Vill	12.334	10.64	-3.24	0.00467
Anxa2	12.284	14.13	3.60	0.02466	Slc25a10	14.554	12.86	-3.24	0.00150
Csf1	9.071	10.92	3.60	0.00661	Gnmt	11.085	9.40	-3.22	0.02214
Tubb2b	8.914	10.76	3.60	0.01092	Paqr9	12.509	10.82	-3.22	0.03481
Lamc2	9.459	11.30	3.59	0.01778	Gm15491	6.210	4.52	-3.22	0.01847
Gna15	4.689	6.53	3.59	0.00274	Bid	12.702	11.02	-3.21	0.00243
Lhpp	7.971	9.81	3.58	0.00363	Slc37a4	13.974	12.29	-3.20	0.00212
Bax	11.253	13.09	3.58	0.03821	Aqp11	11.651	9.98	-3.19	0.00770
Tnfrsf23	5.997	7.84	3.58	0.03558	C330021F23Rik	8.008	6.33	-3.19	0.01256
Tceal3	4.728	6.56	3.57	0.01552	Paqr7	12.411	10.74	-3.19	0.00161
C7	7.895	9.73	3.56	0.01083	Fmo3	6.731	5.06	-3.18	0.00489
Rab39	3.858	5.69	3.56	0.02393	Dmgdh	13.444	11.77	-3.18	0.00111
Ctss	10.246	12.07	3.54	0.01085	Mep1b	14.092	12.42	-3.18	0.01571
Gm10603	4.910	6.74	3.54	0.00974	Sardh	13.160	11.49	-3.18	0.03805
Ccl5	6.311	8.14	3.54	0.02911	Zpld1	8.772	7.10	-3.18	0.00772
Cldn4	9.765	11.59	3.54	0.00229	Neil2	8.331	6.67	-3.17	0.00310
Klrb1b	4.673	6.50	3.54	0.01607	Gcnt1	15.911	14.25	-3.17	0.00781
Ppp1r36	4.762	6.58	3.53	0.00448	Acad10	11.852	10.19	-3.16	0.02995
A430105I19Rik	8.946	10.77	3.53	0.04280	Neurog2	7.258	5.60	-3.14	0.02911
Loxl4	7.154	8.97	3.53	0.00189	Timp3	17.246	15.60	-3.14	0.00198
Rcan1	12.564	14.38	3.53	0.00273	Vps8	12.114	10.47	-3.13	0.00274
Wnt4	7.497	9.31	3.52	0.00484	Pctp	12.434	10.79	-3.13	0.00146
Ncaph	5.986	7.80	3.52	0.00124	Tmem106a	13.076	11.43	-3.13	0.00174
Ascl4	4.127	5.94	3.51	0.03827	Enpp3	12.414	10.77	-3.12	0.00552
Adam8	5.517	7.33	3.51	0.00251	RP23-336I5.8	7.948	6.30	-3.12	0.02545
Camk2b	7.866	9.67	3.50	0.00784	Tprkb	13.896	12.26	-3.11	0.00556
Rrad	6.508	8.31	3.50	0.00157	Cyp51	14.672	13.04	-3.11	0.00178
S100a11	12.081	13.88	3.48	0.00543	B4galt5	13.156	11.52	-3.10	0.00961
Aurka	5.801	7.60	3.47	0.00320	Ahcyl2	14.276	12.65	-3.10	0.03813

Rnaset2b	7.676	9.46	3.45	0.02382	Ggt1	15.970	14.34	-3.09	0.00857
Zfp52	9.117	10.90	3.45	0.00992	Idi1	9.393	7.76	-3.09	0.03862
Galnt12	6.903	8.69	3.45	0.00319	Tmem8	13.820	12.19	-3.09	0.02198
Arg2	9.102	10.89	3.45	0.01678	Slc16a6	9.666	8.04	-3.09	0.00210
Sulf2	9.679	11.46	3.43	0.01607	Tpk1	13.408	11.78	-3.09	0.00174
Arhgap27os1	4.021	5.80	3.43	0.00592	Sec14l3	10.158	8.53	-3.09	0.01607
Kif6	4.621	6.40	3.43	0.02266	P2ry1	8.565	6.95	-3.06	0.00475
Klhdc8a	10.206	11.98	3.42	0.00280	Lrrc8b	12.161	10.55	-3.06	0.03160
Cdca5	5.263	7.03	3.40	0.01367	Mccc1	13.538	11.92	-3.06	0.01408
Krt18	10.045	11.81	3.40	0.01080	Dgat2	13.195	11.58	-3.05	0.00465
Mrc1	7.684	9.44	3.39	0.00892	Gk	15.922	14.31	-3.05	0.00280
2610016A17Rik	5.160	6.92	3.39	0.02995	Gpr137b	12.715	11.11	-3.04	0.00224
8430429K09Rik	8.262	10.02	3.38	0.01110	Gm10658	8.987	7.39	-3.02	0.03055
Nrip2	7.246	9.00	3.36	0.00264	Hgd	15.088	13.49	-3.02	0.00849
H2-Eb1	12.353	14.10	3.36	0.03579	Angptl1	6.039	4.45	-3.02	0.01282
Pcdh9	4.528	6.28	3.36	0.00499	Etv1	10.500	8.91	-3.01	0.01833
Aox3	6.053	7.80	3.35	0.01857	Ggct	12.044	10.45	-3.01	0.01107
Fgf12	5.677	7.42	3.35	0.00822	Gm15651	7.435	5.85	-3.00	0.03732
C1ql3	6.776	8.52	3.35	0.00624	Fah	14.642	13.06	-3.00	0.00400
Ptprr	6.800	8.54	3.35	0.00719	Retsat	13.745	12.16	-3.00	0.00586
Gcsam	4.674	6.41	3.33	0.04308	Tsga10	10.548	8.97	-2.99	0.01857
Plcd4	7.204	8.94	3.33	0.00273	Shmt2	13.903	12.33	-2.98	0.01338
Prrg4	8.897	10.63	3.32	0.03885	Rhov	9.677	8.10	-2.98	0.02515
Dnah1	3.685	5.41	3.31	0.02088	Fam169b	10.044	8.47	-2.97	0.00535
Ctsk	8.084	9.81	3.31	0.00309	Aldh9a1	15.151	13.59	-2.96	0.00273
Fhdc1	6.101	7.82	3.30	0.02306	B230216N24Rik	9.041	7.48	-2.95	0.00400
Dennd2a	7.880	9.60	3.30	0.01372	Acox2	13.075	11.51	-2.95	0.00946
Kcnk1 TWIK-1	11.502	13.22	3.30	0.01210	Fam132a	12.109	10.55	-2.95	0.01085
Casp4	7.303	9.02	3.30	0.03426	Cbs	13.566	12.01	-2.95	0.02584

Il1rl1	5.287	7.01	3.30	0.00326	Slc6a20a	10.481	8.92	-2.94	0.01780
Tmem253	6.009	7.73	3.30	0.00135	Pcca	13.264	11.71	-2.93	0.00442
RP23-110C17.2	7.802	9.52	3.29	0.01079	Ctbs	12.672	11.12	-2.93	0.00120
Slc31a2	10.271	11.99	3.28	0.00257	Ndrg1	16.679	15.13	-2.93	0.04681
Tnc	8.873	10.59	3.28	0.00400	Trpc1	11.606	10.06	-2.93	0.03554
Tnfsf13b	5.614	7.33	3.28	0.00314	Trp53bp2	12.687	11.14	-2.92	0.01214
Nbl1	9.208	10.92	3.28	0.00424	Etv5	9.698	8.16	-2.91	0.02419
Rps6kl1	4.148	5.85	3.26	0.00492	Sms	10.389	8.85	-2.91	0.00434
H2-Aa	12.691	14.40	3.26	0.02155	Miox	16.775	15.24	-2.90	0.00295
Pdpn	8.408	10.11	3.26	0.01376	Zdhhc2	9.119	7.58	-2.90	0.01715
Tubb6	7.924	9.63	3.26	0.00606	RP24-315D19.11	7.676	6.14	-2.90	0.00808
Col18a1	12.105	13.81	3.26	0.01533	Tyw3	11.337	9.81	-2.89	0.01367
Pdgfrl	4.838	6.54	3.25	0.00629	Bphl	14.518	12.99	-2.89	0.00259
Col9a3	3.788	5.49	3.25	0.00508	Slc5a2	14.170	12.64	-2.88	0.02351
Mfge8	12.210	13.91	3.24	0.01003	Bhlhe40	12.484	10.96	-2.88	0.02790
Melk	4.661	6.36	3.24	0.00287	Hmgcr	12.047	10.52	-2.88	0.03426
Phgdh	8.585	10.28	3.24	0.00671	Lrp3	9.669	8.15	-2.87	0.04379
Asf1b	5.770	7.46	3.23	0.01979	Slc2a2	13.459	11.94	-2.87	0.02506
Tlr11	5.218	6.91	3.22	0.02465	Bdnf	9.032	7.51	-2.86	0.02807
Clec12a	7.341	9.03	3.22	0.04123	Map3k15	11.561	10.04	-2.86	0.03310
Hhip	4.725	6.41	3.22	0.00253	Fbp1	16.408	14.89	-2.86	0.01116
Rdh12	5.367	7.05	3.22	0.01119	Ahcy	12.638	11.13	-2.85	0.01108
Sfn	7.820	9.51	3.22	0.00648	Apln	9.266	7.76	-2.85	0.00946
Adgre1	8.227	9.91	3.21	0.00401	Rars2	12.293	10.78	-2.85	0.00275
Igsf10	5.972	7.66	3.21	0.00895	Acad9	12.269	10.76	-2.84	0.00669
Ighm	9.262	10.95	3.21	0.01354	Gm11992	11.627	10.12	-2.83	0.00368
Ihh	9.269	10.95	3.21	0.00201	Glb1l	13.107	11.60	-2.83	0.01468
Ube2c	5.481	7.16	3.21	0.00293	Hps3	11.797	10.30	-2.83	0.01670
Cd68	7.865	9.54	3.20	0.02101	Fmo4	11.370	9.87	-2.83	0.00174

Eno2	5.659	7.34	3.20	0.02202	Cmtm6	14.239	12.74	-2.83	0.01357
Tcf19	7.586	9.26	3.20	0.00552	Nt5dc2	10.384	8.89	-2.83	0.00138
9530052E02Rik	5.124	6.80	3.19	0.01094	Slc25a34	11.381	9.89	-2.82	0.00217
Hspa1b	9.413	11.09	3.19	0.00390	Slc22a2	14.377	12.88	-2.82	0.01278
Tmem158	5.699	7.37	3.19	0.00263	Olfm4	10.885	9.39	-2.81	0.00622
Crybb3	3.941	5.61	3.18	0.03070	Acat1	16.374	14.88	-2.81	0.00163
Pla2g5	9.080	10.74	3.16	0.02049	Btbd11	7.600	6.11	-2.81	0.04314
Ifi27	11.354	13.02	3.16	0.00984	Mpv17l	15.562	14.08	-2.80	0.00401
Fbln1	8.200	9.86	3.16	0.00204	Slc22a18	14.518	13.03	-2.80	0.00717
Lilra5	8.049	9.71	3.16	0.03799	Ivd	15.055	13.57	-2.79	0.01279
Kirrel3	4.804	6.46	3.14	0.00946	Cyp4f15	6.039	4.56	-2.79	0.01071
Fsip1	4.868	6.52	3.13	0.00354	Kcnk5	13.422	11.94	-2.79	0.01521
Muc1	9.403	11.05	3.12	0.00563	Fggy	12.868	11.39	-2.79	0.00207
Cldn6	5.328	6.97	3.12	0.02474	Cyp2j7	7.156	5.68	-2.78	0.01226
Bbc3	6.507	8.15	3.12	0.00568	Ifrd2	12.293	10.82	-2.78	0.00767
Car13	7.968	9.61	3.11	0.01298	St6galnac2	13.368	11.89	-2.78	0.00174
Cpn1	12.017	13.65	3.10	0.02184	Cyp2j9	10.778	9.30	-2.78	0.00356
Lag3	5.174	6.81	3.10	0.01653	Trib2	12.493	11.02	-2.77	0.01716
Serpinb6a	11.206	12.84	3.10	0.01925	Sorcs1	5.843	4.37	-2.77	0.01993
Hpgds	7.737	9.37	3.09	0.00151	Acnat1	9.434	7.97	-2.76	0.01867
Spdl1	6.605	8.23	3.09	0.00138	Adra1a	9.639	8.17	-2.76	0.02501
Tax1bp3	9.408	11.04	3.09	0.02463	Chst7	10.838	9.38	-2.75	0.00356
Epb4.2	6.217	7.84	3.09	0.00318	Grpel2	12.511	11.05	-2.75	0.00257
Sgol2a	5.168	6.80	3.09	0.00801	Slc17a1	15.923	14.46	-2.75	0.01320
Dclk1	5.251	6.88	3.09	0.01348	Cpox	12.601	11.15	-2.74	0.01140
Trip13	4.816	6.44	3.08	0.00433	Gm16170	6.402	4.95	-2.74	0.00563
Clec5a	5.727	7.35	3.07	0.01620	Gm10872	7.301	5.85	-2.74	0.00227
Gm26902	3.806	5.42	3.07	0.01749	Plekhm2	11.596	10.15	-2.73	0.04325
Pld4	8.381	10.00	3.07	0.00171	Gcdh	15.037	13.59	-2.73	0.00365

Ccdc106	6.505	8.12	3.06	0.00983	Fahd2a	12.487	11.04	-2.73	0.01944
Mctp2	7.778	9.39	3.06	0.00720	Aldh1l1	14.502	13.06	-2.73	0.04658
Maff	7.106	8.72	3.05	0.02584	Dcst1	9.927	8.48	-2.72	0.01155
Litaf	10.803	12.41	3.05	0.00094	Zfp101	11.088	9.64	-2.72	0.03104
Cpxm1	7.925	9.53	3.04	0.00257	Gm5475	6.119	4.68	-2.71	0.03335
Kif23	7.580	9.18	3.04	0.04144	Proc	13.381	11.94	-2.71	0.00214
Hap1	7.364	8.97	3.04	0.02161	Amt	12.618	11.18	-2.71	0.00200
Spi1	7.359	8.96	3.03	0.02552	AI464131	10.721	9.29	-2.71	0.04732
Zmat3	10.329	11.92	3.02	0.02515	Gpm6a	13.317	11.89	-2.69	0.03309
Cnp	8.829	10.42	3.02	0.00508	F13b	13.670	12.24	-2.69	0.00235
Slc35g2	7.382	8.98	3.02	0.01730	Neat1	15.388	13.96	-2.69	0.03694
Gpr39	6.965	8.56	3.01	0.00138	Tmem150a	12.510	11.08	-2.68	0.00287
Flt3	5.424	7.01	3.00	0.00400	RP23-327M1.1	8.554	7.13	-2.68	0.03257
Ucp2	11.694	13.28	3.00	0.00326	Phyh	15.935	14.51	-2.68	0.00280
9530052C20Rik	4.063	5.65	3.00	0.01752	Top1mt	10.997	9.57	-2.68	0.00241
Sec14l2	7.190	8.77	3.00	0.00204	D2hgdh	10.946	9.53	-2.68	0.02463
Diap3	4.970	6.55	3.00	0.00492	Aspa	14.360	12.94	-2.68	0.01224
Myrfl	3.371	4.95	3.00	0.04439	Fam214a	11.849	10.43	-2.68	0.02681
Ccdc92	8.092	9.67	2.99	0.00105	Acy1	13.246	11.83	-2.68	0.01638
BC016579	4.434	6.01	2.99	0.00891	Klf15	12.819	11.40	-2.67	0.00464
Trnp1	5.015	6.59	2.99	0.01058	Nipsnap1	13.712	12.29	-2.67	0.00681
Dpt	7.449	9.03	2.99	0.02636	Apoh	9.583	8.17	-2.67	0.03917
Mgl2	8.185	9.76	2.98	0.02101	Cryl1	14.488	13.07	-2.67	0.02641
Cyp4v3	8.768	10.34	2.98	0.00274	Arhgef39	10.209	8.79	-2.67	0.01807
Ncf4	5.980	7.55	2.97	0.03729	Gda	9.585	8.17	-2.66	0.02476
Ifitm1	9.026	10.60	2.97	0.02911	Myom3	8.088	6.68	-2.66	0.00570
Tspan6	9.156	10.72	2.97	0.00789	Fancd2os	8.703	7.29	-2.66	0.00892
Junos	4.991	6.55	2.95	0.01720	Esm1	11.742	10.34	-2.65	0.00459
Lpcat2	6.134	7.70	2.95	0.00605	Tmtc4	10.872	9.47	-2.65	0.00605

Lat2	7.230	8.79	2.94	0.01614	Gpcpd1	11.832	10.43	-2.64	0.01907
Tmem171	9.241	10.80	2.94	0.02047	RP24-448C16.7	10.176	8.78	-2.63	0.01825
Serpine1	7.332	8.89	2.94	0.01062	Herc4	12.988	11.59	-2.63	0.00478
Stx11	6.241	7.79	2.93	0.00395	Mettl1	10.906	9.51	-2.63	0.01684
Aldh1a7	10.096	11.65	2.93	0.00383	Mansc4	9.811	8.42	-2.63	0.01201
Rell2	4.897	6.45	2.93	0.00864	Gatb	12.792	11.40	-2.63	0.00671
Ager	5.294	6.84	2.93	0.00623	Slc5a8	14.732	13.34	-2.63	0.01607
6820408C15Rik	6.127	7.68	2.92	0.02020	Gm17115	7.318	5.93	-2.62	0.00829
1810062G17Rik	5.028	6.58	2.92	0.04215	Slc25a13	13.126	11.74	-2.62	0.00618
Pgf	4.700	6.25	2.92	0.02863	Dbt	13.245	11.86	-2.62	0.00496
Dapp1	7.392	8.93	2.91	0.00171	Amdhd2	12.010	10.62	-2.62	0.00607
Rgs19	8.136	9.67	2.90	0.03614	Gm10644	7.680	6.29	-2.62	0.02171
Slc10a6	5.518	7.05	2.90	0.02339	Tcn2	16.519	15.13	-2.61	0.00698
Cyp4f16	8.739	10.27	2.90	0.00623	Perm1	8.759	7.37	-2.61	0.01125
Smoc2	9.574	11.11	2.89	0.00629	Sephs2	14.578	13.19	-2.61	0.00204
Olfm2	4.656	6.19	2.89	0.02381	Shpk	10.577	9.19	-2.61	0.00259
Ptafr	5.657	7.19	2.89	0.01424	Slc19a1	12.277	10.90	-2.61	0.00679
Lgi3	4.290	5.82	2.88	0.02145	Akr1c21	17.261	15.88	-2.60	0.02342
Sertad1	9.507	11.03	2.88	0.00583	Hnf1aos1	10.218	8.84	-2.60	0.02456
Fcgr1	6.422	7.94	2.87	0.01749	Cyp4a31	13.912	12.54	-2.60	0.01716
Col3a1	11.024	12.55	2.87	0.00274	Sp5	8.879	7.50	-2.60	0.02202
Orai2	7.319	8.84	2.87	0.00677	Grhpr	13.622	12.25	-2.59	0.03095
Ighg2b	7.627	9.14	2.86	0.04379	Pdzd3	12.691	11.32	-2.59	0.00654
Irf7	9.988	11.50	2.85	0.01862	Prkd3	12.742	11.37	-2.58	0.02246
Lamb3	8.181	9.69	2.85	0.00809	BC021785	14.431	13.06	-2.58	0.00295
6720489N17Rik	6.868	8.38	2.85	0.04575	Slc11a2	12.587	11.22	-2.58	0.01648
Tlr4	8.677	10.19	2.85	0.03517	Ighv1-81	7.230	5.87	-2.57	0.01758
Milr1	5.926	7.44	2.85	0.04864	Insig1	11.317	9.96	-2.57	0.00693
Asns	8.381	9.89	2.84	0.01961	Ehhadh	15.573	14.21	-2.56	0.00621

Rhou	7.365	8.87	2.84	0.00359	Tmem88b	8.500	7.14	-2.56	0.01136
Sh2d4b	6.605	8.11	2.84	0.02351	Hoga1	14.023	12.67	-2.56	0.00434
1700024P16Rik	7.380	8.88	2.83	0.00290	Dpyd	12.350	11.00	-2.55	0.01224
4930512B01Rik	4.589	6.09	2.83	0.01367	Selenbp1	14.985	13.64	-2.55	0.00547
Trpv6	6.509	8.01	2.83	0.00347	Cbx2	8.589	7.24	-2.54	0.02593
5430427O19Rik	4.505	6.00	2.82	0.03785	Abcd4	11.119	9.77	-2.54	0.01016
BC039771	6.120	7.61	2.81	0.00801	Susd3	12.106	10.76	-2.54	0.02465
Scara3	4.380	5.87	2.81	0.01004	Tmem237	11.739	10.40	-2.54	0.00552
Clstn2	6.266	7.75	2.81	0.04600	Fcgr2b	12.221	10.88	-2.53	0.00235
Tubb4a	4.855	6.34	2.80	0.04341	Cyp2j5	17.084	15.74	-2.53	0.00552
Liph	4.655	6.14	2.80	0.00817	BC025446	12.184	10.84	-2.53	0.03875
Cd84	6.100	7.59	2.80	0.00807	Ugt8a	13.536	12.20	-2.52	0.02682
C920025E04Rik	6.192	7.68	2.80	0.04244	Sel1l3	11.954	10.62	-2.52	0.04158
2310015A10Rik	8.262	9.74	2.79	0.00400	Ddah1	13.966	12.64	-2.51	0.03614
Fbp2	9.921	11.40	2.79	0.02453	Isoc2a	13.607	12.28	-2.51	0.01010
1700003E16Rik	6.701	8.17	2.78	0.00953	Pex11g	10.775	9.45	-2.51	0.00775
Stil	4.649	6.12	2.77	0.03230	Hus1	12.151	10.83	-2.50	0.02074
Clca3a1	8.701	10.17	2.76	0.00906	D10Wsu102e	11.642	10.32	-2.50	0.01037
Ndc80	5.015	6.48	2.76	0.01720	Hook2	12.145	10.83	-2.50	0.01201
Cd86	7.157	8.62	2.76	0.00946	Sepp1	18.808	17.49	-2.50	0.02517
Hmmr	6.804	8.27	2.75	0.02669	Poli	9.697	8.38	-2.50	0.00397
Arl4c	8.742	10.20	2.75	0.00739	Vwa1	12.004	10.69	-2.49	0.00701
Tgfbi	9.776	11.24	2.75	0.00671	Srr	13.852	12.54	-2.49	0.00289
Mab21l3	6.562	8.02	2.75	0.01201	Hspd1	15.124	13.81	-2.48	0.00395
Vopp1	9.589	11.05	2.75	0.00584	Suclg2	14.583	13.27	-2.48	0.00195
Plk2	10.318	11.78	2.75	0.00376	0610011F06Rik	13.847	12.54	-2.48	0.04765
Lrrc17	5.987	7.44	2.74	0.04280	Zfp810	12.191	10.88	-2.47	0.03588
Fcer1g	8.127	9.58	2.74	0.01241	Cml2	12.283	10.98	-2.47	0.01053
Mmp2	7.701	9.16	2.74	0.00911	Tmem86b	8.740	7.44	-2.47	0.04588

Mok	9.471	10.93	2.74	0.02729	Cfb	9.368	8.06	-2.47	0.02394
Dpysl2	8.198	9.65	2.74	0.00353	Igha	12.875	11.57	-2.47	0.00929
Dtl	5.139	6.59	2.74	0.00606	Sh3bp2	10.387	9.08	-2.47	0.02201
Ccnjl	4.840	6.29	2.73	0.01648	Slc4a4	15.533	14.23	-2.47	0.04569
Hmga1	8.587	10.04	2.73	0.04040	Acadm	16.042	14.74	-2.46	0.01620
Shc4	5.129	6.58	2.73	0.02043	Gnpda1	12.987	11.69	-2.46	0.00538
Pcdhb9	4.920	6.37	2.73	0.01370	Pafah2	13.095	11.80	-2.46	0.01790
Pdzd4	6.029	7.47	2.72	0.00259	Abcb6	6.956	5.67	-2.45	0.00767
Lzic	9.564	11.01	2.72	0.00490	Serpinc1	6.779	5.49	-2.45	0.04339
P2ry14	8.131	9.57	2.72	0.03406	Mapt	13.023	11.73	-2.45	0.03503
Fbn1	9.178	10.62	2.72	0.01981	A1cf	12.670	11.38	-2.45	0.04190
Il3ra	6.311	7.75	2.72	0.02351	Slc27a2	17.943	16.66	-2.44	0.01573
Cd109	5.481	6.92	2.71	0.03943	Hsd17b11	14.922	13.64	-2.44	0.00402
Chaf1b	6.561	8.00	2.71	0.00719	Spats2l	10.414	9.13	-2.44	0.00717
Cldn7	9.527	10.96	2.70	0.02915	Bcat1	12.143	10.86	-2.43	0.02593
Uhrf1	6.348	7.78	2.70	0.01648	Gpr155	10.599	9.32	-2.43	0.00606
Zfp286	6.130	7.56	2.70	0.01857	Apob	14.188	12.91	-2.43	0.03750
Csmd1	5.006	6.44	2.70	0.01980	Gm853	12.334	11.05	-2.43	0.00387
Serping1	11.816	13.25	2.70	0.00715	E230001N04Rik	7.480	6.20	-2.43	0.01010
Mmp3	4.604	6.04	2.70	0.00770	Abhd14a	11.785	10.51	-2.42	0.00975
Spns2	11.573	13.00	2.70	0.00216	Trit1	10.177	8.90	-2.42	0.00605
B4galnt1	8.809	10.23	2.68	0.00805	Csgalnact1	9.910	8.64	-2.41	0.01424
Podxl2	6.179	7.60	2.67	0.01902	Larp4b	13.183	11.92	-2.41	0.04454
Epsti1	7.773	9.19	2.67	0.03769	Suox	12.517	11.25	-2.40	0.01395
Ctxn1	6.822	8.24	2.67	0.00684	Pter	15.461	14.20	-2.40	0.00585
2310058D17Rik	7.733	9.15	2.67	0.01903	Gorasp1	11.224	9.96	-2.40	0.00913
Sytl2	10.398	11.81	2.67	0.02511	Gm15348	9.923	8.66	-2.40	0.01314
Spin2c	5.904	7.32	2.66	0.04735	Sh3bp5	12.478	11.21	-2.40	0.00968
Tpm4	12.838	14.25	2.66	0.01227	Mat2a	15.347	14.09	-2.40	0.00504

Tmeff1	4.596	6.01	2.66	0.02693	Pde9a	11.249	9.99	-2.39	0.01411
Ccdc65	5.133	6.54	2.65	0.01828	Rbpms2	12.304	11.05	-2.39	0.01330
Ptp4a1	9.151	10.56	2.65	0.02877	RP23-93F11.4	7.831	6.57	-2.39	0.02879
Cercam	5.085	6.49	2.65	0.01226	4833439L19Rik	16.129	14.87	-2.39	0.00871
Nat14	7.522	8.93	2.65	0.03884	Nr1h4	13.651	12.40	-2.39	0.00671
Nkd1	8.107	9.51	2.64	0.00304	Ttc39c	12.154	10.90	-2.38	0.00697
Ppfia4	7.204	8.60	2.64	0.00866	Ddc	12.332	11.08	-2.38	0.00484
Maged2	10.211	11.61	2.64	0.00506	Klhl8	9.755	8.50	-2.38	0.02116
Mmp19	5.320	6.72	2.64	0.03245	Gm15318	10.782	9.53	-2.37	0.00812
Aldh18a1	7.301	8.70	2.64	0.00913	Pkd2l2	6.867	5.62	-2.37	0.01262
P2ry6	6.378	7.77	2.63	0.00390	Prdm5	8.770	7.52	-2.37	0.00655
Myo15	4.308	5.70	2.63	0.01474	Sdc2	13.463	12.22	-2.37	0.00396
Plk5	6.001	7.39	2.63	0.02969	Hsd3b3	13.272	12.03	-2.37	0.01907
Amica1	5.021	6.41	2.62	0.00710	Gphn	12.416	11.17	-2.37	0.00857
Was	6.035	7.42	2.62	0.01286	Arhgap42	13.080	11.84	-2.37	0.01465
Dnase2a	9.845	11.23	2.62	0.03256	Teddm2	6.331	5.09	-2.36	0.01008
Pacsin1	4.436	5.82	2.61	0.03565	Pop1	9.858	8.62	-2.36	0.01925
Mag	4.745	6.13	2.61	0.01242	Fgl1	6.912	5.68	-2.36	0.00586
Kcne1l	5.344	6.73	2.61	0.00525	1600014C10Rik	12.318	11.08	-2.35	0.00287
Sh3bgrl3	10.997	12.38	2.60	0.03889	Hyi	12.294	11.06	-2.35	0.01051
Sgol1	5.050	6.43	2.60	0.01871	9130023H24Rik	9.260	8.03	-2.35	0.00607
Racgap1	6.822	8.20	2.60	0.00654	2900005J15Rik	7.146	5.92	-2.34	0.04970
Cd74	13.090	14.47	2.59	0.00189	Map10	9.038	7.81	-2.34	0.00536
Camkk1	6.676	8.05	2.59	0.04485	Plekhb2	14.762	13.54	-2.34	0.00821
Bcam	12.169	13.54	2.59	0.01424	Abhd14b	14.727	13.50	-2.34	0.00551
Cpne8	8.583	9.95	2.59	0.00720	Hsd3b2	13.990	12.77	-2.33	0.01833
Sla	7.600	8.97	2.59	0.01348	Gm15459	6.892	5.67	-2.33	0.01688
Gm6030	4.575	5.94	2.58	0.02815	Pipox	15.036	13.81	-2.33	0.00807
Rpusd1	8.005	9.37	2.58	0.01850	Gm6652	7.572	6.35	-2.33	0.01646

Lrriq1	6.207	7.58	2.58	0.04507	Rufy3	13.263	12.04	-2.33	0.02506
Scml2	4.723	6.09	2.58	0.04742	Stra6l	9.923	8.71	-2.33	0.01982
Kif22	5.912	7.28	2.58	0.01226	Tsku	10.794	9.58	-2.32	0.02415
Dcn	11.410	12.78	2.58	0.02446	Bend7	10.727	9.51	-2.32	0.01894
Kif27	6.968	8.33	2.58	0.03921	Slc35a3	13.870	12.66	-2.32	0.02177
Efna4	7.272	8.64	2.58	0.00356	Slco3a1	13.465	12.25	-2.32	0.02959
F2rl1	10.324	11.69	2.57	0.01868	Gm13421	6.452	5.24	-2.31	0.03598
Nrcam	5.773	7.14	2.57	0.00957	Depdc7	11.652	10.44	-2.31	0.00657
H2-K1	13.496	14.86	2.57	0.01687	Gm27216	10.631	9.42	-2.31	0.03967
Lrguk	4.676	6.04	2.57	0.03507	4933427G17Rik	6.912	5.70	-2.31	0.01778
9530077C05Rik	7.309	8.67	2.57	0.00652	Arl6ip1	15.830	14.62	-2.31	0.00715
Catsper4	4.661	6.02	2.56	0.01593	Gcat	11.685	10.48	-2.31	0.01857
Scn7a	8.368	9.73	2.56	0.01631	Edem1	12.986	11.78	-2.31	0.01766
Siglech	4.753	6.11	2.56	0.02465	Slc25a15	13.782	12.58	-2.31	0.00648
Pqlc3	8.026	9.38	2.56	0.00606	Dpep1	13.328	12.12	-2.31	0.01386
Kcnip2	5.471	6.83	2.56	0.01510	Trim7	13.422	12.22	-2.30	0.01119
Ptrf	10.453	11.81	2.55	0.00287	Cat	16.149	14.95	-2.30	0.00639
Stc2	6.722	8.07	2.55	0.01054	Arhgef10l	11.805	10.60	-2.30	0.02685
Ifi204	8.382	9.73	2.55	0.03589	Tspan7	10.487	9.29	-2.29	0.00963
Gm17501	6.234	7.58	2.55	0.04310	Slc44a4	12.375	11.18	-2.29	0.04962
Fst	8.319	9.67	2.55	0.01270	Gpx3	20.747	19.55	-2.29	0.02584
A930001C03Rik	7.429	8.78	2.55	0.02544	Oxct1	16.804	15.61	-2.28	0.02463
Scel	9.719	11.07	2.55	0.01766	Ganc	11.369	10.18	-2.28	0.02966
Lgals3bp	11.255	12.60	2.54	0.01982	Fbxo7	12.706	11.52	-2.28	0.00473
Nfkbid	5.423	6.77	2.54	0.02465	Agmo	8.803	7.62	-2.27	0.01195
Bmp1	8.746	10.09	2.54	0.02043	Galns	13.108	11.92	-2.27	0.03012
Hpse	4.866	6.21	2.54	0.02415	Gm20400	8.198	7.01	-2.27	0.03409
Gpr173	5.455	6.79	2.53	0.01519	Scrn3	12.610	11.43	-2.27	0.01092
Hrasls	7.200	8.54	2.53	0.00784	Tbxas1	10.330	9.15	-2.27	0.00520

Upk3b	8.319	9.66	2.53	0.02351	Atad3a	11.944	10.76	-2.27	0.01825
Trf	5.786	7.12	2.53	0.04751	Tiparp	11.894	10.71	-2.27	0.02667
Igf2bp1	8.179	9.52	2.53	0.02000	Acaa1b	13.416	12.24	-2.27	0.00829
Plxdc1	4.483	5.82	2.52	0.01676	Pth1r	15.009	13.83	-2.27	0.00483
Gm26826	4.546	5.88	2.52	0.04308	C1qtnf3	14.106	12.93	-2.27	0.00603
Cdc6	4.667	6.00	2.52	0.04990	Slc35d2	11.006	9.83	-2.26	0.02120
Abcc3	6.689	8.02	2.52	0.03534	C130074G19Rik	14.496	13.32	-2.25	0.04664
Anxa5	12.731	14.07	2.52	0.00601	BC035947	12.791	11.62	-2.25	0.02630
Rec8	9.038	10.37	2.52	0.02959	Rabggta	12.107	10.94	-2.25	0.01668
Pnck	5.329	6.66	2.51	0.01676	Nol10	10.324	9.16	-2.25	0.01085
1700102P08Rik	4.546	5.87	2.51	0.03999	Afm	11.227	10.06	-2.25	0.00913
Dok1	7.651	8.98	2.51	0.03959	Esrp2	11.227	10.06	-2.24	0.02875
Slc7a4	7.925	9.25	2.51	0.03552	Lrpap1	15.287	14.12	-2.24	0.02351
Mlkl	7.268	8.59	2.50	0.01866	Tmem220	9.514	8.35	-2.24	0.00381
Sgcd	4.748	6.07	2.50	0.04945	Slc35f3	10.200	9.04	-2.24	0.02534
Cx3cr1	8.343	9.66	2.49	0.00401	Afap1l1	12.363	11.20	-2.24	0.00825
Mybl1	7.013	8.33	2.49	0.02838	Lbr	11.466	10.31	-2.23	0.01381
Slc7a6	8.466	9.78	2.49	0.00273	Cep85	11.487	10.33	-2.23	0.01426
Igsf6	6.500	7.81	2.49	0.04698	Ddo	11.588	10.44	-2.22	0.01381
S1pr2	7.762	9.08	2.49	0.00216	Abcd3	14.921	13.77	-2.22	0.00659
Gm20939	7.504	8.82	2.48	0.04152	Fastkd1	11.312	10.16	-2.22	0.02196
Basp1	6.549	7.86	2.48	0.03894	Gpd1	14.711	13.56	-2.22	0.01336
Nfe2l3	5.619	6.93	2.48	0.02306	Sfxn1	14.261	13.11	-2.21	0.00619
Clu	13.479	14.79	2.48	0.01970	Gpt	10.493	9.35	-2.21	0.04673
Aldh1b1	6.914	8.22	2.47	0.00526	Zc3h12d	7.096	5.95	-2.21	0.04465
Rasl11b	10.043	11.35	2.47	0.00497	Acp6	12.026	10.88	-2.21	0.01729
Rras	9.757	11.06	2.47	0.02916	Me1	15.376	14.24	-2.21	0.00848
Abhd8	9.653	10.96	2.47	0.01239	Rmnd1	11.641	10.50	-2.20	0.01041
Mb21d1	5.592	6.89	2.47	0.01126	Cda	13.468	12.33	-2.20	0.03413

Gm684	5.323	6.62	2.47	0.01388	Vegfa	13.983	12.85	-2.20	0.02968
Gm11734	4.688	5.99	2.46	0.03079	3110057O12Rik	10.797	9.66	-2.20	0.01386
Ncf1	7.235	8.54	2.46	0.01053	Tulp3	12.008	10.87	-2.20	0.02351
Tor3a	9.833	11.13	2.46	0.00484	Mut	14.282	13.15	-2.19	0.01404
Itgb6	12.203	13.50	2.46	0.01004	Erlin1	13.473	12.34	-2.19	0.00585
Tubb2a	10.047	11.35	2.46	0.01424	Acpp	9.120	7.99	-2.19	0.04134
Plet1	10.959	12.26	2.45	0.00444	Catsper2	7.723	6.59	-2.19	0.00822
Cd72	8.327	9.62	2.45	0.02534	Sfxn5	10.611	9.48	-2.19	0.04560
Lyl1	5.930	7.22	2.45	0.01427	B3gat2	11.608	10.48	-2.19	0.03292
Shisa4	8.856	10.15	2.45	0.01119	Rcl1	10.383	9.26	-2.18	0.00671
Lrp2bp	5.272	6.56	2.45	0.01201	Lrrc75b	8.874	7.75	-2.18	0.03513
Ifitm10	5.046	6.33	2.44	0.01961	Gm16861	6.758	5.63	-2.18	0.03061
Lypd6b	8.124	9.41	2.44	0.04666	Btnl9	10.588	9.46	-2.18	0.01640
Dnaaf3	4.927	6.21	2.44	0.04882	Rpgrip1	7.767	6.64	-2.18	0.02959
Cmtm3	9.723	11.01	2.44	0.04310	Apeh	13.518	12.40	-2.17	0.03554
Arpc1b	11.223	12.50	2.43	0.02848	0610043K17Rik	8.166	7.05	-2.17	0.00911
Nudt18	9.394	10.67	2.42	0.02296	Akr1d1	12.503	11.39	-2.17	0.02145
Spag5	9.048	10.32	2.42	0.03037	Snhg17	9.826	8.71	-2.17	0.02593
Epcam	12.369	13.65	2.42	0.00921	Nudt19	16.135	15.02	-2.17	0.01094
Fblim1	9.505	10.78	2.42	0.00426	Nkain1	8.762	7.65	-2.16	0.01841
Tnfsf9	5.293	6.57	2.42	0.04490	Ghr	16.297	15.18	-2.16	0.01240
RP23-480P21.2	6.322	7.60	2.42	0.00800	Atf7ip2	6.486	5.37	-2.16	0.02012
Slc38a2	12.044	13.32	2.42	0.04575	Abhd17c	12.624	11.51	-2.16	0.02959
Lrrc25	5.474	6.74	2.41	0.04685	Arhgap18	12.753	11.64	-2.16	0.00990
Lynx1	10.212	11.48	2.41	0.00606	Pitpnc1	12.084	10.98	-2.16	0.04583
Ywhah	11.738	13.00	2.41	0.01600	Irx3	10.711	9.61	-2.15	0.01928
Tcaf2	7.579	8.85	2.41	0.00433	4933431E20Rik	11.440	10.33	-2.15	0.02689
C3ar1	7.183	8.45	2.41	0.00518	Slc16a4	13.744	12.64	-2.15	0.02465
Cd44	7.398	8.66	2.41	0.01126	Abhd1	8.771	7.67	-2.15	0.02178

Kcnk2	7.285	8.55	2.40	0.04430	Cdcp1	11.821	10.72	-2.15	0.03070
Lrrc73	4.393	5.66	2.40	0.04727	Atp6v1b2	14.756	13.66	-2.14	0.02526
Pxdc1	7.612	8.88	2.40	0.00534	Nadsyn1	10.932	9.83	-2.14	0.02872
Npdc1	10.931	12.19	2.40	0.01724	Erc2	11.131	10.03	-2.14	0.00989
Siglece	6.210	7.47	2.40	0.00387	Def8	13.845	12.75	-2.14	0.01311
Aplp1	7.576	8.84	2.40	0.04787	N4bp2l1	9.528	8.43	-2.14	0.01676
Relt	4.470	5.73	2.39	0.02351	Invs	10.937	9.84	-2.13	0.03731
Hoxb9	10.830	12.09	2.39	0.00492	Dhfr	13.311	12.22	-2.13	0.04444
Osr2	9.051	10.31	2.39	0.03186	Rffl	11.641	10.55	-2.13	0.02163
Samd14	7.247	8.51	2.39	0.02371	Treh	13.009	11.92	-2.13	0.02351
Nipal2	8.925	10.18	2.39	0.00984	Aasdh	9.914	8.83	-2.13	0.03634
Susd1	8.582	9.84	2.39	0.01062	Sgpp1	13.610	12.52	-2.12	0.01629
Hs3st1	6.949	8.20	2.39	0.04505	Igsf11	11.774	10.69	-2.12	0.01395
1700112E06Rik	5.803	7.05	2.38	0.02679	Galnt14	11.936	10.85	-2.12	0.01861
Cdh11	9.574	10.83	2.38	0.01353	Mfsd9	11.295	10.21	-2.12	0.01376
Serpinf1	7.063	8.31	2.38	0.04949	3110083C13Rik	6.456	5.37	-2.12	0.03256
Fut4	6.622	7.87	2.38	0.01043	Dpf3	7.670	6.59	-2.11	0.00719
Tekt2	5.500	6.75	2.38	0.03634	2310016G11Rik	6.808	5.73	-2.11	0.02685
Mmp9	4.597	5.85	2.38	0.01730	Gm32569	6.250	5.17	-2.11	0.01894
4833422C13Rik	6.740	7.99	2.38	0.02083	Slc38a4	8.068	6.99	-2.11	0.04537
Hoxa4	6.204	7.45	2.37	0.01159	Pisd-ps1	12.246	11.17	-2.11	0.02198
Cdkl3	8.296	9.54	2.37	0.01083	Nt5dc1	11.841	10.76	-2.11	0.01004
Bak1	9.782	11.03	2.37	0.01720	Tmem175	11.206	10.14	-2.10	0.01607
Ggt7	5.552	6.80	2.37	0.04267	Rhbdd2	11.711	10.64	-2.10	0.02342
Mpeg1	10.478	11.72	2.37	0.00873	Ugt2b37	14.372	13.31	-2.09	0.00669
Tlr7	6.942	8.18	2.36	0.04111	Slc3a1	15.648	14.58	-2.09	0.01367
Ubxn11	7.223	8.46	2.36	0.00703	Clpx	13.444	12.38	-2.09	0.02177
Lxn	7.954	9.19	2.36	0.02549	Idh2	14.974	13.91	-2.09	0.00606
Efna2	4.963	6.20	2.36	0.03932	Dph1	9.783	8.72	-2.09	0.02517

Serpinb9	11.188	12.43	2.36	0.02057	Slc16a13	11.467	10.41	-2.09	0.02101
Mmp14	8.810	10.05	2.36	0.00870	Nkx3-1	6.236	5.18	-2.08	0.04840
Tes	10.670	11.91	2.36	0.01618	Slc25a16	13.728	12.67	-2.08	0.01599
Ccdc102a	6.974	8.21	2.36	0.03070	Sfxn2	11.652	10.60	-2.07	0.02306
Bub1b	6.100	7.34	2.36	0.01282	Csad	14.164	13.11	-2.07	0.02171
Rras2	11.376	12.61	2.35	0.01056	Gatsl3	9.808	8.76	-2.07	0.02116
Cd33	6.331	7.56	2.35	0.00847	Nudt12	12.106	11.06	-2.07	0.02018
Runx1	6.600	7.83	2.35	0.01291	Smim10l2a	8.531	7.48	-2.07	0.02641
Relb	7.784	9.02	2.35	0.01607	BC065397	7.116	6.07	-2.07	0.02966
Reep2	5.002	6.23	2.35	0.01329	Smpd2	12.382	11.34	-2.06	0.02973
Trim59	7.612	8.84	2.34	0.02045	Frs3	9.708	8.66	-2.06	0.03921
Serpinb6b	9.602	10.82	2.33	0.03543	Hsd17b7	10.898	9.86	-2.06	0.03652
Irak3	6.622	7.84	2.33	0.01367	Ccdc163	9.554	8.51	-2.06	0.00606
Rhbdl2	6.684	7.90	2.33	0.00475	Pip5k1b	8.287	7.25	-2.06	0.00962
Agt	11.127	12.35	2.33	0.01528	Rundc3b	6.405	5.37	-2.05	0.02537
A930024E05Rik	4.836	6.05	2.33	0.03882	Cutc	10.142	9.10	-2.05	0.00637
Tacstd2	10.356	11.57	2.32	0.00323	Mest	7.015	5.98	-2.05	0.04454
Ormdl3	10.463	11.68	2.32	0.00565	Bckdha	14.359	13.32	-2.05	0.02908
Hk2	4.912	6.13	2.32	0.01996	Zfp39	9.102	8.07	-2.04	0.01244
Ier3	10.667	11.88	2.32	0.01982	Ptcd3	11.733	10.70	-2.04	0.01367
Vav1	6.311	7.52	2.32	0.02705	Slc7a2	9.121	8.10	-2.04	0.04735
Rgs6	9.463	10.67	2.31	0.01212	Harbi1	10.345	9.32	-2.03	0.01828
Rundc3a	8.346	9.56	2.31	0.01220	Hibch	12.884	11.87	-2.02	0.01894
Ppp1r3b	6.614	7.82	2.31	0.00829	Tmem178	11.570	10.55	-2.02	0.00841
Rbm3	11.139	12.35	2.31	0.01736	Pisd-ps2	9.716	8.70	-2.02	0.02436
Mfap2	6.486	7.69	2.31	0.01062	B230369F24Rik	7.485	6.47	-2.02	0.03070
Unc13d	5.829	7.03	2.31	0.03409	Slc22a4	12.455	11.44	-2.02	0.02057
Tmem98	8.639	9.84	2.30	0.01116	Pdzk1	16.507	15.50	-2.01	0.01469
Slc22a15	7.742	8.95	2.30	0.00806	1700055D18Rik	7.235	6.23	-2.01	0.01381

6030419C18Rik	5.578	6.78	2.30	0.01638	Gucd1	13.616	12.61	-2.01	0.00891
Arhgap27os2	7.716	8.92	2.30	0.02794	Fbxl21	8.672	7.67	-2.01	0.01573
Wdr6	10.603	11.80	2.30	0.03889	Neu1	15.279	14.28	-2.01	0.01687
Asrgl1	9.281	10.48	2.30	0.00720	Upp2	11.657	10.65	-2.00	0.02441
Ccnb2	6.233	7.43	2.30	0.00898	Slc47a1	15.574	14.57	-2.00	0.01468
Npy1r	8.420	9.62	2.29	0.04215					
Cftr	9.219	10.42	2.29	0.03104					
Htr2b	5.707	6.90	2.29	0.03673					
Catsperd	5.091	6.29	2.29	0.02386					
Rad18	7.081	8.28	2.29	0.01016					
Slc11a1	7.180	8.37	2.29	0.00898					
Clec4a1	5.451	6.64	2.29	0.03921					
Arhgef25	8.298	9.49	2.29	0.00901					
Stap2	9.391	10.58	2.28	0.01656					
Hspa1a	8.407	9.59	2.27	0.00715					
Tmem71	6.682	7.86	2.27	0.02269					
Arhgap8	7.115	8.30	2.27	0.00709					
Pirb	7.077	8.26	2.27	0.02463					
Cnn2	10.766	11.94	2.26	0.02115					
Cpne7	7.036	8.21	2.26	0.01565					
Tmem173	7.652	8.83	2.26	0.00685					
Laptm5	9.256	10.43	2.26	0.01276					
sept.04	10.791	11.97	2.26	0.02074					
Mvp	10.513	11.69	2.26	0.03921					
Gm9949	6.051	7.22	2.26	0.04262					
Tmem100	6.467	7.64	2.25	0.03525					
Arhgap22	8.464	9.63	2.25	0.00361					
Cacnb3	8.948	10.12	2.25	0.01291					
Gm16685	6.906	8.07	2.25	0.02417					

Osmr	9.365	10.53	2.25	0.01278
C030037D09Rik	7.497	8.66	2.25	0.00947
Dusp10	5.945	7.11	2.24	0.04991
Tlr13	6.834	8.00	2.24	0.04990
Col1a2	11.035	12.20	2.24	0.04085
Zbtb42	7.933	9.10	2.24	0.01875
Cd1d1	7.390	8.55	2.24	0.01766
Tspyl3	5.615	6.78	2.24	0.03002
Efemp2	8.855	10.02	2.24	0.00733
Gm13270	5.370	6.53	2.24	0.01996
Slc16a3	5.625	6.79	2.24	0.01648
Rnf32	7.520	8.68	2.23	0.02973
Wnt7b	9.026	10.19	2.23	0.01010
B3galt1	5.799	6.96	2.23	0.03367
C1ra	9.413	10.57	2.23	0.01877
Sphk1	8.339	9.49	2.22	0.01766
Ttc22	7.456	8.61	2.22	0.01320
Pax8	12.122	13.27	2.22	0.01132
AI429214	7.975	9.12	2.22	0.04370
F420014N23Rik	5.794	6.94	2.22	0.04659
C3	8.843	9.99	2.22	0.02979
Ncmap	4.580	5.73	2.21	0.03731
Hfe	10.497	11.64	2.21	0.00537
Fam105a	7.965	9.11	2.21	0.01437
Dab1	8.147	9.29	2.21	0.02139
Loxl1	8.886	10.03	2.21	0.01946
St14	11.339	12.48	2.21	0.01857
Elf3	9.617	10.76	2.20	0.00782
Btla	6.452	7.59	2.20	0.01861

Stac3	5.108	6.25	2.20	0.02202
Plcxd1	7.819	8.96	2.20	0.02185
Jund	11.113	12.25	2.20	0.02228
Slc44a3	9.247	10.39	2.20	0.00637
Fam131a	7.164	8.30	2.20	0.03592
C5ar2	4.271	5.41	2.20	0.04310
Uba52	5.524	6.66	2.20	0.01444
Lrrc8e	6.032	7.16	2.19	0.01664
Zak	9.298	10.43	2.19	0.04927
Tnfrsf19	6.736	7.87	2.19	0.04787
Lrrc27	6.636	7.77	2.19	0.01841
Ppp1r15a	10.284	11.41	2.19	0.01431
Tubg2	5.651	6.78	2.19	0.01894
Jdp2	8.792	9.92	2.19	0.04542
Pkib	6.219	7.35	2.18	0.04023
Slc9a9	6.561	7.69	2.18	0.00584
Atf5	9.465	10.59	2.18	0.02243
Gpr65	6.734	7.86	2.18	0.01828
Lrrc20	7.591	8.71	2.18	0.00934
Crlf2	7.131	8.25	2.17	0.04486
Stc1	8.049	9.17	2.17	0.04441
Gm19426	5.182	6.29	2.16	0.03428
P3h4	8.089	9.20	2.16	0.04379
Mr1	9.163	10.28	2.16	0.00906
Atp8b5	6.620	7.73	2.16	0.01000
Colec12	10.549	11.66	2.16	0.01419
Slc15a1	4.956	6.07	2.16	0.04404
Gas7	8.120	9.23	2.16	0.03503
Styk1	7.110	8.22	2.16	0.02410

Ptger4	7.251	8.36	2.15	0.03367
Nfam1	6.548	7.65	2.15	0.01925
Cd4	5.720	6.82	2.15	0.03406
B3gnt9	6.914	8.02	2.15	0.03136
Fzd3	6.950	8.05	2.15	0.02233
Gm13648	7.495	8.60	2.14	0.01881
Mns1	4.982	6.08	2.14	0.02584
Dbf4	8.096	9.20	2.14	0.01079
Fam124a	8.175	9.27	2.14	0.01083
Rgs14	5.565	6.66	2.14	0.02351
Art4	8.867	9.96	2.13	0.03311
Me2	8.860	9.95	2.13	0.01058
Clrn3	12.615	13.70	2.13	0.01638
Zfp599	6.010	7.10	2.12	0.01648
Trim45	6.297	7.38	2.12	0.00586
Pvr	9.512	10.59	2.12	0.01045
Cxcl10	7.949	9.03	2.12	0.02346
Lrrc34	5.281	6.36	2.12	0.01314
Aqp2	12.913	13.99	2.12	0.01497
Selplg	7.474	8.55	2.11	0.01248
Apobr	6.283	7.36	2.11	0.02956
Cdc20	6.441	7.52	2.11	0.02233
Dusp5	6.777	7.85	2.11	0.04215
9430091E24Rik	6.556	7.63	2.11	0.03832
Lgals12	4.953	6.03	2.10	0.01902
C1qtnf6	7.575	8.65	2.10	0.01381
Hyls1	7.065	8.14	2.10	0.02962
Gsdmc3	7.500	8.57	2.10	0.03577
Scrn1	7.848	8.92	2.10	0.02205

Tpm1	12.988	14.06	2.10	0.04379
Cd81	14.171	15.24	2.09	0.01600
Cdh3	8.118	9.18	2.09	0.03766
Parvg	6.820	7.88	2.09	0.03884
Gm13420	6.191	7.25	2.09	0.03311
Ggt5	7.846	8.90	2.08	0.02145
Rcan2	9.796	10.85	2.08	0.03552
Tinagl1	11.492	12.54	2.07	0.00605
Serinc2	9.940	10.99	2.07	0.03579
Cpne2	8.648	9.70	2.07	0.01125
Gfra4	5.683	6.73	2.07	0.01564
Tmem119	9.160	10.20	2.06	0.03802
Fermt3	7.483	8.53	2.06	0.03010
Ric3	7.927	8.97	2.06	0.02635
Gm13205	5.712	6.75	2.05	0.02331
Larp6	6.266	7.30	2.05	0.02091
Sepn1	8.106	9.14	2.05	0.02353
Spr-ps1	7.201	8.24	2.05	0.04379
1810055G02Rik	8.692	9.73	2.05	0.01334
Spon1	10.593	11.62	2.04	0.00838
Pamr1	8.629	9.66	2.04	0.01381
Ccl25	8.999	10.03	2.04	0.04892
Pstpip1	6.591	7.62	2.04	0.02916
Serpinh1	12.365	13.39	2.03	0.01025
Ifit3b	8.724	9.75	2.03	0.01986
Fstl3	8.629	9.65	2.03	0.02342
Sass6	7.170	8.19	2.03	0.04666
Slc35e4	8.500	9.52	2.02	0.01706
Gm1976	7.789	8.80	2.02	0.01004

Adamtsl2	9.204	10.22	2.02	0.01687
Efemp1	10.781	11.79	2.02	0.01638
Cfh	13.952	14.96	2.01	0.03515
Vstm4	9.289	10.30	2.01	0.04310
Lbp	7.976	8.99	2.01	0.02594
Cpb2	6.816	7.82	2.01	0.01511
Rab31	10.553	11.56	2.01	0.01975
Tspan17	8.748	9.75	2.01	0.04432
Marveld3	9.902	10.90	2.00	0.01887
Bag2	10.403	11.40	2.00	0.02797

Supplemental Table 2. Micro-computed tomography of L5 vertebrae from Control and cKO mice

MALES	**Control (n=5)**	**cKO (n=5)**	***p*-value**
BMD (mg*cm^{-3})	208 ± 35	126 ± 12	**0.001**
BV/TV (%)	18.94 ± 4.37	7.20 ± 1.66	**0.001**
Tb.Th (mm)	0.055 ± 0.004	0.048 ± 0.003	**0.015**
Tb.N (mm^{-1})	3.39 ± 0.55	1.49 ± 0.26	**0.0001**
Tb.Sp (mm)	0.19 ± 0.02	0.27 ± 0.03	**0.002**

FEMALES	**Control (n=5)**	**cKO (n=7)**	***p*-value**
BMD (mg*cm^{-3})	115 ± 27	82 ± 43	0.165
BV/TV (%)	11.54 ± 2.52	7.14 ± 3.77	**0.048**
Tb.Th (mm)	0.059 ± 0.003	0.052 ± 0.007	0.067
Tb.N (mm^{-1})	1.94 ± 0.35	1.33 ± 0.67	0.095
Tb.Sp (mm)	0.29 ± 0.04	0.35 ± 0.11	0.209

BMD, bone mineral density; BV/TV, bone volume per total volume;
Tb.Th, trabecular thickness; Tb.N, trabecular number; Tb.Sp, trabecular separation;
Data are means ± SD. The experiment was performed on 3.5-month old mice,
28 days after the end of DOX treatment. Statistical analysis was performed by the unpaired Student's t-test.

Supplemental Table 3. Micro-computed tomography of femora from Control and cKO mice.

MALES		**Control (n=5)**	**cKO (n=5)**	***p*-value**
distal metaphysis trabecular 3D analyses				
	BMD (mg*cm^{-3})	139 ± 35	138 ± 19	0.971
	BV/TV (%)	11.5 ± 3.8	11.67 ± 2.18	0.933
	Tb.Th (mm)	0.058 ± 0.003	0.064 ± 0.003	**0.011**
	Tb.N (mm^{-1})	1.95 ± 0.57	1.82 ± 0.27	0.64
	Tb.Sp (mm)	0.23 ± 0.03	0.28 ± 0.02	**0.021**
cortical 3D analyses				
mid-shaft	**Ct.Th (mm)**	0.20 ± 0.01	0.18 ± 0.02	0.052
	TMD (mg*cm^{-3})	1175 ± 22	1123 ± 20	**0.004**
distal diametaphysis	**Ct.Th (mm)**	0.16 ± 0.01	0.14 ± 0.02	**0.041**
	TMD (mg*cm^{-3})	1056 ± 15	926 ± 30	**<0.0001**
distal metaphysis	**Ct.Th (mm)**	0.13 ± 0.01	0.11 ± 0.01	**0.017**
	TMD (mg*cm^{-3})	912 ± 16	810 ± 18	**<0.001**
cortical 2D analyses				
mid-shaft	**T.Ar (mm^2)**	0.82 ± 0.05	0.73 ± 0.06	**0.022**
	Ct.Ar (mm^2)	0.82 ± 0.05	0.72 ± 0.06	**0.023**
	Ct.Ar/T.Ar (%)	99.4 ± 0.1	99.5 ± 0.1	0.248
distal diametaphysis	**T.Ar (mm^2)**	0.85 ± 0.06	0.75 ± 0.1	0.086
	Ct.Ar (mm^2)	0.84 ± 0.05	0.74 ± 0.1	0.076
	Ct.Ar/T.Ar (%)	98.6 ± 0.3	97.9 ± 0.5	**0.03**

distal metaphysis	**T.Ar (mm^2)**	0.77 ± 0.03	0.63 ± 0.07	**0.003**
	Ct.Ar (mm^2)	0.75 ± 0.03	0.6 ± 0.07	**0.002**
	Ct.Ar/T.Ar (%)	96.7 ± 0.3	96.2 ± 0.5	0.069

FEMALES		**Control (n=4)**	**cKO (n=7)**	***p*-value**
distal metaphysis trabecular 3D analyses				
	BMD ($mg*cm^{-3}$)	80 ± 18	115 ± 45	0.174
	BV/TV (%)	6.24 ± 1.27	9.8 ± 4.8	0.188
	Tb.Th (mm)	0.064 ± 0.004	0.067 ± 0.006	0.31
	Tb.N (mm^{-1})	0.99 ± 0.24	1.43 ± 0.62	0.204
	Tb.Sp (mm)	0.37 ± 0.06	0.35 ± 0.07	0.669
cortical 3D analyses				
mid-shaft	**Ct.Th (mm)**	0.20 ± 0.01	0.19 ± 0.02	0.155
	TMD ($mg*cm^{-3}$)	1198 ± 20	1163 ± 43	0.169
distal diametaphysis	**Ct.Th (mm)**	0.19 ± 0.02	0.17 ± 0.02	0.124
	TMD ($mg*cm^{-3}$)	1113 ± 56	1021 ± 73	0.058
distal metaphysis	**Ct.Th (mm)**	0.15 ± 0.01	0.13 ± 0.02	0.116
	TMD ($mg*cm^{-3}$)	932 ± 26	851 ± 69	0.052
cortical 2D analyses				
mid-shaft	**T.Ar (mm^2)**	0.79 ± 0.06	0.71 ± 0.09	0.143
	Ct.Ar (mm^2)	0.79 ± 0.06	0.70 ± 0.09	0.132
	Ct.Ar/T.Ar (%)	99.5 ± 0.2	99.2 ± 0.2	0.08
distal diametaphysis	**T.Ar (mm^2)**	0.90 ± 0.07	0.79 ± 0.13	0.183

	Ct.Ar (mm^2)	0.89 ± 0.07	0.78 ± 0.13	0.154
	Ct.Ar/T.Ar (%)	99.1 ± 0.5	98.1 ± 0.5	**0.013**
distal metaphysis	**T.Ar (mm^2)**	0.87 ± 0.09	0.72 ± 0.19	0.17
	Ct.Ar (mm^2)	0.84 ± 0.09	0.68 ± 0.19	0.146
	Ct.Ar/T.Ar (%)	96.8 ± 0.3	94.6 ± 2.3	0.089

BMD, bone mineral density; BV/TV, bone volume per total volume; Tb.Th, trabecular thickness;
Tb.N, trabecular number; Tb.Sp, trabecular separation; Conn.D, connectivity density; Ct.Th, cortical thickness;
TMD, tissue mineral density; T.Ar, total area; Ct.Ar, cortical bone area.
Data are means ± SD. The experiment was performed on 3.5-month old mice,
28 days after the end of DOX treatment. Statistical analysis was performed by the unpaired Student's t-test.

Supplemental Table 4. Histomorphometric analysis of vertebrae from male Control and cKO mice.

	Control (n=4)	cKO (n=4)	*p*-value
BV/TV (%)	22.58 ± 4.27	18.94 ± 2.58	0.205
Tb.Th (um)	43.96 ± 3.63	34.03 ± 4.22	0.161
Tb.N (mm^{-1})	5.11 ± 0.69	4.74 ± 0.39	0.402
Tb.Sp (um)	154 ± 33	172 ± 19	0.409
Ob.S/B.Pm (%)	12.84 ± 6.08	11.02 ± 3.17	0.618
N.Ob/B.Pm (mm^{-1})	11.16 ± 4.29	10.57 ± 3.17	0.832
OS/BS (%)	1.62 ± 0.52	31.08 ± 8.20	**0.006**
O.Th (um)	2.88 ± 0.52	6.18 ± 0.61	**0.0002**
OV/BV (%)	0.21 ± 0.01	9.70 ± 0.02	**0.003**
Oc.S/B.Pm (%)	7.34 ± 0.47	9.80 ± 3.05	0.209
N.Oc/B.Pm (mm^{-1})	6.33 ± 0.60	8.73 ± 1.00	**0.009**

BV/TV, bone volume per total volume; Tb.Th, trabecular thickness; Tb.N,trabecular number; Tb.Sp, trabecular separation;
Ob.S/B.Pm, osteoblast surface per bone perimeter; N.Ob/B.Pm, number of osteoblasts per bone perimeter;
OS/BS, osteoid surface per bone surface; O.Th, osteoid thickness, Oc.S/B.Pm, osteoclast surface per bone perimeter;
N.Oc/B.Pm, number of osteoclasts per bone perimeter; OV/BV, osteoid volume per bone volume.
Data are means ± SD. The experiment was performed on 3.5-month old mice,
28 days after the end of DOX treatment. Statistical analysis was performed by the unpaired Student's t-test.

Supplementary Methods

Animals

Xpr1 null mice Mice with gene trap mutated *Xpr1* allele were obtained from Lexicon (TF0891 mutants).

Conditional knockout of Xpr1 in the nephron. Mice with floxed exon 2 of *Xpr1* were obtained from Cyagen. The procedures used to generate the *Xpr1*$^{lox/lox}$-Pax8-rtTA/LC-1 mice were described by Traykova-Brauch et al. (2008, *Nat Med,* 14**:** 979-984).

The animals were maintained *ad libitum* on the standard laboratory chow diet (KLIBA NAFAG diet 3800). The conditional inactivation of *Xpr1* in the nephron was induced by 2-weeks treatment with doxycycline (DOX, 2 mg/ml in drinking water) of 8-weeks old *Xpr1*$^{lox/lox}$-Pax8-rtTA/LC-1 mice (cKO mice). In parallel the same DOX treatment was provided to their littermate controls Xpr1$^{lox/lox}$ mice (Control mice).

Microdissection

Mice were anesthetized with ketamine/xylazine and perfused with 10 ml of DMEM + 40 µg/ml liberase (Roche). The left kidneys were then decapsulated and cut into small pieces that were incubated for 30 minutes at 37°C in DMEM + 40 µg/ml liberase. Kidney pieces were washed 2 times with DMEM and kidney segments were microdissected in ice-cold 0.05% BSA/DMEM.

RNA extraction and quantitative PCR

Half frozen kidneys were homogenized with a Polytron homogenizer in D-buffer (4 M Guanidium thiocyanate, 25 mM Na-citrate, 0.5% Na-lauroylsarcosyl, 0.1 M β-mercaptoethanol), NaOAc (pH 4.0), saturated phenol (pH 4.0) and chloroform-isomylalcohol solution and then centrifuged at 10'000 g for 20 min. Isopropanol was added at the aqueous phase to precipitate the RNA and the RNA pellet was washed with 70% ethanol and purified with RNeasy Micro Kit (QIAGEN). Reverse transcription was performed with 1 µg of RNA using PrimeScript RT Reagent kit (TAKARA). 4 µl of cDNA was used for quantitative real-time PCR to assess Xpr1 mRNA expression. Assays were performed with Taqman probes (Applied Biosystems Mm01284709_m1) and master mix (Applied Biosystems). mRNA expression was normalized with GAPDH expression.

Western blot

Half decapsulated kidneys were homogenized with a polytron in 3 ml RIPA buffer (20 mM Tris-HCl (pH 7.2), 150 mM NaCl, 0.1% SDS, 0.5% Na-deoxycholate, 1% Triton-X-100, protease inhibitors). Protein extracts were sonicated and centrifuged for 10 minutes at 10,000

g. The supernatant was recovered and protein concentration measured with Pierce BCA protein assay reagent (Thermo) and then adjusted to 8 mg/ml with RIPA buffer. Samples were mixed with Laemmli sample buffer (60 mM Tris-Hcl (pH 6.8), 2% SDS, 10% glycerol, 5% β-mercaptoethanol, 0.01% bromophenol blue) and heated at 95°C for 5 minutes. 40 µg of protein were loaded in Mini-PROTEAN TGX gels, 4-20% (BIO-RAD), and then transferred to nitrocellulose membranes. Membranes were blocked with 5% skim milk in TBST (Tris-buffered saline, 0.1% Tween) for 1 hour at room temperature and then incubated overnight with the primary anti-Xpr1 antibody (Proteintech) in the blocking solution, at 4°C. Membranes were washed and incubated with anti-rabbit horseradish peroxidase conjugated IgG in 5% skim milk in TBST for 1 hour at room temperature. After the washing steps, SuperSignal west dura extended duration substrate (Thermo) was used and signal visualized on Kodak Biomax XAR film (Kodak). Of note, we also tested the anti-XPR1 antibodies from Genetex (GTX108458), Abcam (ab118315), Origene (TA308700), Abcam (ab88911) as well as four different home-made antibodies. All these antibodies were nonspecific in both Western blot and IHC applications.

Metabolic cages and urine and blood analyses

Mice were housed individually in metabolic cages (Tecniplast) with free access to food and water and were habituated for 2-3 days before urine collection. Plasma and urine sodium and potassium concentrations were determined by flame photometry (Instrumentation laboratory). Urinary pH was measured by using a pH meter (Metrohm). Plasma and urine creatinine, glucose, calcium and magnesium concentrations were measured in the Laboratoire Central de Chimie Clinique, Centre Hospitalier Universitaire Vaudoise (CHUV) University Hospital (Lausanne, Switzerland). Plasma and urine osmolality was measured with osmometer from Advanced Instruments (Model 2020). Urinary phosphate concentration was determined using Malachite Green Phosphate Assay kit (BioAssay Systems) following manufacturer's instructions. Plasma phosphate was measured as described above on plasma samples recovered by tail incision. Urinary amino acids were measured by BIOCRATES Life Sciences (Innsbruck, Austria). ELISA kits were used according to manufacturer's instructions: FGF23 (Kainos Japan CY-4000), PTH (Immutopics 60-2305), CTX-I (RatLaps AC-0671), osteocalcin (Bioquote by Biomedical Technologies BT-470), TRAP (antibodies-online GmbH ABIN627521), 1,25(OH)2-vitamin D3 EIA (ImmunoDiagnostic Systems AC-62F1), and alkaline phosphatase activity colorimetric kit (Abcam ab83369).

GFR measurement

GFR was determined from FITC coupled inulin clearance on anesthetized mice as described previously (Qi et al, AJP Renal, 286(3):F590-6, 2004). FITC-inulin (5% in 0.85% NaCl) was dialyzed overnight and 50 µl of dialyzed FITC-inulin was injected retro-orbitally. Venous blood was collected from the saphenous vein 3, 7, 10, 15, 20, 40 and 60 minutes after FITC-

inulin injection. Plasma was diluted 5 times with 0.5 M HEPES and measured for the fluorescence intensity by Nanodrop 3300 (Thermo).

Texas-Red albumin injection

Anesthetized mice were intravenously injected with 10 μg/g body weight of TR-albumin (Rockland Immunochemicals) dissolved in 0.9% NaCl. After 5 minutes, the kidneys were fixed by retrograde perfusion through the abdominal aorta with 2% paraformaldehyde. After an overnight incubation in 2% PFA, the kidneys were embedded in paraffin and sections of 3 μm were performed.

Primary culture of proximal tubular cells

Primary cultures of proximal tubular cells from DOX-untreated Control and cKO male mice were prepared according to the method described by Terryn et al. (Am. J. Physiol, 2007 293(2): F467-85). Briefly, renal cortex was cut into ~1-mm^3 cubes in ice-cold dissection solution (NaCl 137 mM, KCl 5.4 mM, Na_2HPO_4 0.25 mM, glucose 10 mM, KH_2PO_4 0.44 mM, $CaCl_2$ 1.3 mM, $MgSO_4$ 1 mM, $MgCl_2$ 0.5 mM, glycine 5 mM, alanine 1 mM, HEPES 15 mM, pH 7.4). The ~1-mm^3 cortex cubes were digested with Liberase (50 μg/ml, Sigma) for 30 min. Supernatant was passed through a 100 μm sieve and fragments longer than 100 μm were collected into dissection solution containing 1% BSA. After centrifugation, proximal tubules were suspended into a culture medium (1:1 DMEM/F12 supplemented with FCS 1%, HEPES 15 mM pH 7.2, insulin 5 μg/ml, transferrin 5 μg/ml, selenium 50 nM, hydrocortisone 50 nm, penicillin 100 U/ml and streptomycin 100 mg/ml) and seeded onto collagen-coated 48-well plates. Tubules were left unstirred 48 h at 37°C and then the medium was changed every 2 days. After 7 days of culture, cells were treated with 5 mg/ml of doxycycline for 24 hours. Then, cells were washed out of doxycycline and used for Pi or glucose uptake/efflux experiments.

Phosphate and glucose uptake and efflux

Phosphate uptake and efflux were measured according to the method of Giovannini et al. (Cell Rep. 2013 27;3(6):1866-73). For the **uptake**, cells were washed with a transport solution without phosphate (NaCl 145 mM, HEPES 10 mM, KCl 5 mM, $CaCl_2$ 2.5 mM, $MgSO_4$ 1.8 mM, glucose 5 mM) for 10 min. The same solution with [^{33}P]phosphate (0.5 μCi/ml) was then added. After different times of incubation (10, 20 and 30 min), cells were washed 3 times with ice-cold PBS and lysed in Triton X-100 1%. Intracellular [^{33}P]phosphate was determined by scintillation counting and normalized to the protein content which was determined by BCA protein assay (ThermoFisher). Phosphate **efflux** was determined after 30 min of uptake. Cells were washed 3 times with the transport solution and incubated in the same solution but with 10 mM phosphate added. 50 μl were collected at different time points (10, 20, 30 min and 60 min) and radioactivity was measured by scintillation counting. Percentage of phosphate efflux was calculated as the ratio of released phosphate to initial intracellular phosphate after 30 min of uptake. Intracellular [^{33}P]phosphate was also determined at the end of the efflux. Glucose efflux was determined on cells loaded with

[^{14}C]α-methyl-D-glucopyranoside (1 mCi/ml) in the incubation medium in which glucose was replaced by α-methyl-D-glucopyranoside (2 mM). After 15 min of uptake, cells were washed with a cold solution and efflux was followed during 3, 6 and 15 min.

Phosphate uptake and efflux from freshly isolated renal tubules. Renal tubules were isolated according to the method described for primary culture experiments (see above) at the exception that (i) the whole kidney was used for tubules isolation, (ii) the kidney pieces were digested with collagenase type 1A (Sigma) and, (iii) that the tubular suspension was sieved trough a 40 μm sieve. Microscope examination of final tubular suspensions revealed more than 80% proximal tubules. The Pi uptake was measured at 37°C in a transport solution containing NaCl 145 mMol, Hepes 10 mMol, mannitol 10 mMol, KCl 5 mMol, $MgSO_4$ 1.8 mMol, Na_2HPO_4 50 μMol, pH 7.4 completed with [^{33}P]phosphate (0.1 μCi/ml). At the end of 30 min uptake tubules were centrifuged at 1000 g, resuspended in ice-cold [^{33}P]phosphate-free transport solution and filtered through 8 μm MF membranes (Millipore). After washing with ice-cold transport solution the filters were counted in a scintillation counter. For Pi efflux the centrifuged tubules were resusupended in the Pi-free transport solution completed with phosphonoformic acid (PFA) 5mMol. Tubules were centrifuged 3, 8 or 40 min after the beginning of the efflux time-course, resusupended in ice cold transport solution, filtered through 8 μm MF membranes and counted in a scintillation counter. Background was determined by 5 seconds incubation of tubules in [^{33}P]phosphate-containing transport solution at 4°C, followed by filtration of tubular suspensions through 8 μm MF membranes and counting in a scintillation counter.

Micro-computed tomography

For ex-vivo micro-computed tomography analysis, femurs were scanned on a SkyScan 1076 machine (Skyscan, Kontich, Belgium) in 70% ethanol with voxel size 18μm, filter AI 0.5mm, exposure 1180 ms, voltage 63kV and current 166 μA. 3D reconstructions were visualized by CTVol Version 2.1 (Bruker). Bone mineral density (BMD) was measured in reference to 0.25 and 0.75 g/cm3 calcium phosphate standards with 2mm diameter (Skyscan). Images were reconstructed using NRecon Version 1.6.9.3 (SkyScan) and analyzed by CTAn Version 1.13.2.1 (SkyScan 2003-11, Bruker 2012-13). Sections 0.5 to 1.5mm from distal growth plate for metaphyseal bone, 2.15-2.58 mm for diametaphyseal bone and 0.45 mm mid-shaft calculated from distal growth plate and minor trochanter underwent automated segmentation into cancellous and cortical bone with grayscale thresholds of 80/255 and 85/255. Vertebral body L5 was assessed in interpolated regions of interest between 3 manually selected elliptic planes confined to the trabecular area. Morphometry was obtained from binarized images using 3D techniques for all parameters, except for cortical total area and cortical bone area for which 2D morphometry techniques were used.

Bone histomorphometry

Processing of samples, staining, and histomorphometric analysis of bone specimen was performed by the bone histomorphometry core facility of Prof. Roland Baron at Harvard Dental School, Boston, MA, USA. Vertebrae were dissected, fixed with PFA 4% - PBS overnight, rinsed with tap water overnight, washed in ethanol 70%, dehydrated in acetone and embedded in methyl methacrylate. Using a microtome (RM2255, Leica, Germany), 4 μm frontal plane sections were cut, and consecutive sections were stained with Von Kossa and 2% Toluidine Blue (pH3.7). Another consecutive section was stained with TRAP and counterstained with Toluidine Blue. Images were obtained using Nikon E800 microscope and Olympus DP71 camera. Image analysis was performed using Olympus CellSens software at 20X magnification or otherwise specified by scale bar. Histomorphometric data was obtained at 200X magnification in a 1.8 mm high x 1.3 mm wide region 200 μm from the growth plate using OsteoMeasure software (Osteometrics Inc., Decatur, GA, USA). Structural parameters bone volume (BV/TV), trabecular thickness (Tb.Th), trabecular number (Tb.N) and trabecular separation (Tb.Sp) were obtained from means of 2 consecutive sections.