

Table S1. Immunoassay information for blood and serum chemistry, muscle adenosine triphosphate content as well as muscle and adipose tissue protein analysis.

<u>Blood & Serum chemistry</u>	<u>Assay information</u>
Bicarbonate & BUN	VetScan Comprehensive Diagnostic Profile, Abaxis, 500-0038
Creatinine	LC-MS/MS method
25(OH)D ₃	IDS, AC-35F1
1,25(OH) ₂ D ₃	IDS, AC-62F1

<u>Muscle & adipose tissue</u>	<u>Assay information</u>
ATP Assay Kit	Abcam, ab83355
Mouse UCP1 ELISA kits	Aviva Systems Biology, OKCD02970
Mouse UCP3 ELISA kits	Aviva Systems Biology, OKEH05259

Table S2. PCR primer information.

Gene	Forward primer	Reverse primer
Ankrd2	TGGACATGCTAGTGCTAGAGG	CGCTTTCTGCTTGCCTTT
Atf3	GAGGATTGCTAACCTGACACC	TTGACGGTAACTGACTCCAGC
Atrogin-1	CAGCTCGTGAGCGACCTC	GGCAGTCGAGAAGTCCAGTC
Cidea	TGACATTCATGGGATTGAGAC	GGCCAGTTGTATGACTAAAGAC
CD137	CGTCAGAACTCCTGTGATAAC	GTCCACCTATGCTGGAGAAGG
Cox2	AACCCAGGGGATCGAGTGT	CGCAGCTCAGTGTGTTGGGAT
Csrp3	GGGGGAGGTGAAAATGTG	CAGGCCATGCACTGGAAACA
Cyfip2	ATGACCACCCACGTCACTTG	CCTGCTCTGAAGTTCGTGTC
Dio2	AATTATGCCTCGGAGAAGACCG	GGCAGTTGCCTAGTGAAGGT
Fhl1	GAETGCCGCAAGCCCATAA	CCAAGGGTGAAGGCACCTT
Fos	TTGAGCGATCATCCGGTC	GCGTAGTCCATACTGGCAAG
IL1β	GCAACTGTTCTGAACTCAACT	ATCTTTGGGGTCCGTCAACT
IL6	TAGTCCTCCTACCCAAATTCC	TTGGTCTTAGCCACTCCTTC
Ly6a	AGGAGGCAGCAGTTATTGTGG	CGTTGACCTTAGTACCCAGGA
Mup1	GAAGCTAGTTCTACGGGAAGGA	AGGCCAGGATAATAGTATGCCA
Murf-1	GTGTGAGGTGCCTACTTGTC	GCTAGTCTCTGTCCCTTGGA
Myd88	TCATGTTCTCCATACCCCTGGT	AAACTGCGAGTGGGGTCAG
Myl2	ATCGACAAGAATGACCTAAGGG	ATTTTCACGTTCACTCGTCCT
Myl3	TGGGGAAAGCCAAAACAGGAAG	AGCCATCAGTTCTACCTCA
Myod	CCACTCCGGGACATAGACTTG	AAAAGCGCAGGTCTGGTGAN
Myogenin	GAGACATCCCCCTATTCTACCA	GCTAGTCCGCTCATAGCC
Myostatin	AGTGGATCTAAATGAGGGCAGT	GTTCAGGCGCAGCTTAC
Pax7	TCTCCAAGATTCTGTGCCGAT	CGGGGTTCTCTCTTATACTCC
Pdk4	AGGGAGGTCGAGCTGTTCTC	GGAGTGTCACTAACGGTCA
Pgc1α	TATGGAGTGACATAGAGTGTGCT	GTCGCTACACCACTTCAATCC
Pgf2α synthase	CTGGACTCATCGCAAACACAA	AGGAAGCCTTGACTTCTGTCTA
Ppargc1α	AGAGCCCCATCTGCTCTCTC	ACTGGTAGTCTGCAAAACCAAA
Prdm16	CCCCACATTCCGCTGTGAT	CTCGCAATCCTGCACTCA
Sell	TACATTGCCAAAAGCCCTTAT	CATCGTCCATTCCCAGAGTC
SIn	CTTGGGTGGATACCAACGTCTG	CTTCTGCTTACAGCCATAGCC
Sncg	AAAGACCAAGCAGGGAGTAACG	GACCACGATGTTTCAGCCTC
Spp1	AGGAAGAAGTAGGCATTCTGGT	TCGGCTCTGCAATGTTGTCTG
Tbx1	CTGTGGGACGAGTTCAATCAG	TTGTCTACAGGGCACAAAG
Tbc1d1	TTCTGGGGGTGAGTCTCAG	GCAGGGCATTACGGTAGGAG
Tlr2	GCAACCGCTTCTGCTCAG	AGGCCTCTCCCTCTATTGTATT
Tmem26	TTCTGTTGCATTCCCTGGTC	GCCGGAGAAAGCCATTG
TNF-α	CCCTCACACTCAGATCATCTTCT	GCTACGACGTGGGCTACAG
Tnncl	GCGGTAGAACAGTTGACAGAG	CCAGCTCTGGTGTGAT
Tnni1	ATGCCGGAAGTTGAGAGGAA	TCCGAGAGGTAACGCACCTT
Tpm3	ACCACCATCGAGGCGGTAA	CCCTTCCCGCATCATCA
Traf6	AAAGCGAGAGATTCTTCCCTG	ACTGGGACAATTCACTAGAGC
Gapdh	AGGTGGTGTGAACGGATTG	TGTAGACCATGTAGTTGAGGTCA
(internal control)		

Table S3. Serum and blood chemistry of mice. Twelve-month-old *Ctns*^{-/-} mice and WT mice were treated with 25(OH)D₃ (25 µg/kg/day), 1,25(OH)₂D₃ (20 ng/kg/day) or vehicle control (ethylene glycol) for six weeks. All mice were fed *ad libitum*. Data are expressed as mean ± SEM. Results of *Ctns*^{-/-} + Vehicle, *Ctns*^{-/-} + 25(OH)D₃ and *Ctns*^{-/-} + 1,25(OH)₂D₃ mice were compared to those of WT + Vehicle mice, respectively. ^a *p* < 0.05, significantly different in *Ctns*^{-/-} mice than WT mice. ^b *p* < 0.05, significantly different in *Ctns*^{-/-} + 25(OH)D₃ or *Ctns*^{-/-} + 1,25(OH)₂D₃ mice versus *Ctns*^{-/-} + Vehicle mice. BUN, blood urea nitrogen.

	WT + Vehicle (n = 6)	<i>Ctns</i> ^{-/-} + Vehicle (n = 5)	<i>Ctns</i> ^{-/-} + 25(OH)D ₃ (n = 6)	<i>Ctns</i> ^{-/-} + 1,25(OH) ₂ D ₃ (n = 6)
BUN (mg/dL)	25.3 ± 3.8	74.3 ± 15.8 ^a	78.4 ± 21.4 ^a	69.5 ± 11.4 ^a
Creatinine (mg/dL)	0.11 ± 0.03	0.26 ± 0.04 ^a	0.24 ± 0.06 ^a	0.27 ± 0.09 ^a
Bicarbonate (mmol/L)	26.5 ± 2.7	26.8 ± 3.2	26.8 ± 2.3	27.1 ± 2.2
25(OH)D ₃ (ng/mL)	103.5 ± 22.5	38.5 ± 6.3 ^a	55.6 ± 6.8 ^{a,b}	43.2 ± 5.3 ^a
1,25(OH) ₂ D ₃ (pg/mL)	255.6 ± 35.4	98.6 ± 25.4 ^a	113.1 ± 14.6 ^a	158.4 ± 11.3 ^{a,b}

Table S4. Serum and blood chemistry of mice. Twelve-month-old *Ctns*^{-/-} mice and WT mice were treated with 25(OH)D₃ (50 µg/kg/day), 1,25(OH)₂D₃ (40 ng/kg/day) or vehicle control (ethylene glycol) for six weeks. All mice were fed *ad libitum*. Results are expressed and analyzed as in Supplemental Table S3. ^a *p* < 0.05, significantly different in *Ctns*^{-/-} mice than WT mice. ^b *p* < 0.05, significantly different in *Ctns*^{-/-} + 25(OH)D₃ or *Ctns*^{-/-} + 1,25(OH)₂D₃ mice versus *Ctns*^{-/-} + Vehicle mice.

	WT + Vehicle (n = 6)	<i>Ctns</i> ^{-/-} + Vehicle (n = 5)	<i>Ctns</i> ^{-/-} + 25(OH)D ₃ (n = 6)	<i>Ctns</i> ^{-/-} + 1,25(OH) ₂ D ₃ (n = 6)
BUN (mg/dL)	25.3 ± 3.8	74.3 ± 15.8 ^a	78.4 ± 21.4 ^a	69.5 ± 11.4 ^a
Creatinine (mg/dL)	0.11 ± 0.03	0.26 ± 0.04 ^a	0.24 ± 0.06 ^a	0.27 ± 0.09 ^a
Bicarbonate (mmol/L)	26.5 ± 2.7	26.8 ± 3.2	26.8 ± 2.3	27.1 ± 2.2
25(OH)D ₃ (ng/mL)	103.5 ± 22.5	38.5 ± 6.3 ^a	55.6 ± 6.8 ^{a,b}	43.2 ± 5.3 ^a
1,25(OH) ₂ D ₃ (pg/mL)	255.6 ± 35.4	98.6 ± 25.4 ^a	113.1 ± 14.6 ^a	158.4 ± 11.3 ^{a,b}

Table S5. Serum and blood chemistry of mice. Twelve-month-old *Ctns*^{-/-} mice and WT mice were treated with 25(OH)D₃ (75 µg/kg/day), 1,25(OH)₂D₃ (60 ng/kg/day), or vehicle control (ethylene glycol) for six weeks. All mice were fed *ad libitum*. Results are expressed and analyzed as in Supplemental Table S3. ^a *p* < 0.05, significantly different in *Ctns*^{-/-} mice than WT mice. ^b *p* < 0.05, significantly different in *Ctns*^{-/-} + 25(OH)D₃ or *Ctns*^{-/-} + 1,25(OH)₂D₃ mice versus *Ctns*^{-/-} + Vehicle mice.

	WT + Vehicle (n = 4)	<i>Ctns</i> ^{-/-} + Vehicle (n = 4)	<i>Ctns</i> ^{-/-} + 25(OH)D ₃ (n = 6)	<i>Ctns</i> ^{-/-} + 1,25(OH) ₂ D ₃ (n = 6)
BUN (mg/dL)	23.5 ± 6.9	57.8 ± 5.3 ^a	67.8 ± 4.8 ^a	74.3 ± 11.1 ^a
Creatinine (mg/dL)	0.11 ± 0.03	0.26 ± 0.04 ^a	0.28 ± 0.06 ^a	0.29 ± 0.07 ^a
Bicarbonate (mmol/L)	27.4 ± 2.1	27.5 ± 1.6	27.3 ± 14	26.5 ± 2.2
25(OH)D ₃ (ng/mL)	109.6 ± 17.8	38.7 ± 5.9 ^a	114.3 ± 14.3 ^b	53.4 ± 11.5 ^a
1,25(OH) ₂ D ₃ (pg/mL)	274.3 ± 17.5	115.3 ± 21.4 ^a	201.7 ± 21.5 ^a	265.1 ± 11.8 ^b