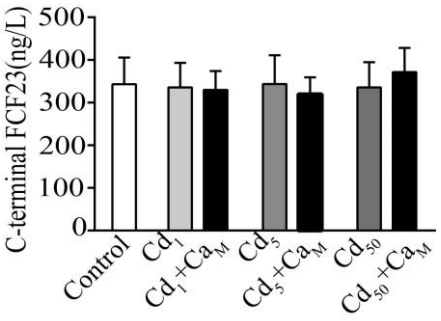

Figure S1: Serum C-terminal FCF23 in the study of moderate Ca supplementation exposure to different diet Cd.

Supplementary Table S1 Composition of AIN-93G diets (g/kg)

Ingredient (g/kg)	
Casein	200
Cornstarch	397.486
Dextrin	132
Sucrose	100
Soybean oil	70
Cellulose	50
Mineral mix[a]	35
Vitamin mix[b]	10
L-Cystine	3
Choline bitartrate	2.5
Tert-Butylhydroquinone	0.014

a The composition of mineral mixture was based on AIN-93G. It provided (mg/kg diet): Ca, 5000; P,3000; Mg, 513; Fe, 45; Zn, 38 and Cu, 6.

b The composition of vitamin mixture was based on AIN-93. The diets contain 1 IU Vitamin D3 per 1g diet

Supplementary Table S2 Primer sequences for quantitative real-time polymerase chain reaction.

	Genes	primers
NM_017008.4	R-GAPDH-S	CTGGAGAAACCTGCCAAGTATG
	R-GAPDH-A	GGTGGAAGAATGGGAGTTGCT
NM_031336.1	R-Klotho -S	TCCGACGAGGACTCTTCTATGT
	R- Klotho -A	GGCTACAACCCCGTCTACTTTA
NM_024146.1	R- Fgfr1-S	GGAGGCTACAAGGTTCGTTACG
	R- Fgfr1-A	AATGCTCCCATACTCGTTCTCC
NM_053763.1	R-Cyp27b1-S	CGGGACACTTCGCACAGTTTAT
	R-Cyp27b1-A	TTCTTCACCATCCGCCGTTAG
NM_013030.1	R-Napi2a-S	TCACAGTCTCATTCGGATTTGGT
	R-Napi2a-A	TGACGAGGAGGATAAGACAAGTGC
NM_201635.3	R-Cyp24a1-S	AATGAGGTCTTGGCTGACTTTCT

	R-Cyp24a1-A	TCACCATCATCTTCCCAAACG
NM_053304.1	R-Colla1(RZ）-S	AGAGGCATAAAGGGTCATCGTG
	R-Colla1(RZ)-A	AGACCGTTGAGTCCATCTTTGC
NM_013004.1	R-Phex-S	CCAAACCACTGCTCCACATCTT
	R-Phex-A	TATGCTCATTGGATGCCTTGTC
NM_001278483.1	R-Runx2(RZ)-S	GCGGACGAGGCAAGAGTTTC
	R-Runx2(RZ)-A	GACTTGGTGCTGAGTTCAGGGAG
NM_053650.1	R-ALP（RZ）-S	GGATCAAAGCAGCATCTTACCAG
	R-ALP（RZ）-A	GCTTTCCCATCTTCCGACACT
NM_012881.2	R-OPN(rz)-S	GATGAACAGTATCCCGATGCCA
	R-OPN(rz)-A	GTCTTCCCGTTGCTGTCCTGA
NM_001037632	R- Osterix-S	CTGGGAAAAGGAGGCACAAAGA
	R- Osterix-A	GGGGAAAGGGTGGGTAGTCATT
NM_013414.1	R- OCN（RZ）-S	AGGGCAGTAAGGTGGTGAATAGA
	R- OCN（RZ）-A	GAAGCCAATGTGGTCCGCTA
NM_203493.3	R-Dmp1(RZ)-S	CGCCCATGGCAAATAGTGAC
	R- Dmp1 (RZ)-A	CTCCTTATCGGCGTCCATCC
NM_130754.1	R-FGF23(RZ)-S	AGAGCCTATTCAGACACTTCCCC
	R-FGF23(RZ)-A	AGCCAGCATCCTCTGATGTGAT
NM_030584.1	R-Sost(RZ)-S	CAACCAGACCATGAACCGGG
	R-Sost(RZ)-A	TCACGAAGCGGGTGTAGTG
NM_001106350.1	R-Dkk1-S	GTGTTCTGGTGGACAGTTTGCC
	R-Dkk1-A	TCTCAATCTCGTCTGGGCTGAT
NM_012870.2	R-OPG-S	GTGTTCTGGTGGACAGTTTGCC
	R-OPG-A	TCTCAATCTCGTCTGGGCTGAT
NM_057149.1	R- RANKL -S	CATCGGGTTCCCATAAAGTCAGT
	R- RANKL -A	GCAAATGTTGGCGTACAGGTAAT
NM_012669.1	R-Tcf1-S	CAGCGGAGGTCCCTTAGTCA
	R-Tcf1-A	CAGACCAATGACCAGGGTAGAG
NM_024355.1	R-Axin2(RZ)-S	ATGTCCATGACAGACAGTAGCGTAG
	R-Axin2(RZ)-A	TGGGTTCTCGGGAAGTGAGG
NM_001105714.1	R-Wnt1(rz)-S	TGGGTTTCTGCTACGTTGCTACT
	R-Wnt1(rz)-A	CAGCCTCGGTTGACGATCTT
NM_053357.2	R-β-catenin-S	TGGTGAAAATGCTTGGGTCG
	R-β-catenin-A	TCTGAAGGCAGTCTGTCGTAATAG

Supplementary Table S3 Summary of food consumption data (g/week) in the 90-day oral toxicity study (mean ± SD, n =10).

	Control	Cd_1	Cd_1+ Ca_L	Cd_1+Ca_M	Cd_1+Ca_H
Week1(day1-7)	111.34±8.67	118.41±2.03	104.44±1.48	111.62±3.5	119.88±3.14
Week2(day8-14)	111.24±2 3.59	128.02±1 9.35	109.62±2 9.29	114.3±15 .94	133.06± 16.29
Week3(day15-21)	135.98±7 .17	144.61±1 4.52	110.58±7 .95*	129.37±9 .09	148.2±1 4.62
Week4(day22-28)	136.64±0 .3	139.86±7 .82	127.89±1 .84*	124.21±6 .53	141.02± 9.85
Week5(day29-35)	124.81±2 .8	136.26±1 2.14	118.76±0 .85*	117.67±7 .53	132.44± 3.17
Week6(day36-42)	120.54±3 .25	141.93±9 .48*	121.38±2 .8	123.62±8 .19	132.41± 9.41*
Week7(day43-49)	123.79±1 0.04	138.11±2 2.81	116.52±1 5.05	120.06±1 3.51	120.77± 17.83
Week8(day50-56)	123.93±1 0.88	141.4±15 .05	123.48±5 .53	126.95±6 .6	131.39± 1.25
Week9(day57-63)	134.82±1 4.46	137.94±9 .04	123.3±0. 18	126.42±4 .43	130.9±1 .03
Week10(d	126.95±4	134.51±2	123.09±5	125.9±2.	130.38±

ay64-70)	.76	.99	.94*	25	3.36
Week11(d	129.47±2	122.03±1	120.38±2	127.55±2	129.64±
ay71-77)	4.15	8.6	2.41	3.95	23.85
Week12(d	113.12±2	102±0.59	133.39±8	123.03±2	124.66±
ay78-84)	0.22		.45	3.8	19.74
Week13(d	101.75±1	99.6±0.5	107.63±7	108.48±2	107.59±
ay85-90)	7.8	1	.49	1	17.42

* Statistically significant from control, $p<0.05$.

Supplementary Table S4 Absolute organ weight and relative organ weight in the 90-day oral toxicity study (mean ± SD, n = 10).

	Control	Cd_1	Cd_1+ Ca_L	Cd_1+ Ca_M	Cd_1+ Ca_H
Body weight[a] (g)	308±52	338±59	310±28	299±27	315±30
Heart(g)	1.01±0.1	0.98±0.2	0.96±0.19	0.92±0.18	0.97±0.23
Liver	8.77±1.24	9.18±2.25	8.11±0.77	7.96±0.79	7.89±0.73
Spleen	0.5±0.12	0.53±0.09	0.54±0.13	0.51±0.14	0.48±0.1
Kidney(paired)	2.14±0.4	2.05±0.42	1.88±0.23	1.88±0.15	2.03±0.28

Uterus	0.55±0.24	0.55±0.16	0.57±0.29	0.64±0.39	0.63±0.17
Ovary(paired)	0.13±0.05	0.2±0.19	0.13±0.02	0.14±0.02	0.13±0.02
Heart/body weight ratios(mg/100 g)	0.33±0.05	0.3±0.06	0.31±0.04	0.31±0.04	0.31±0.06
Liver/body weight ratios(mg/100 g)	2.88±0.46	2.74±0.4	2.62±0.22	2.65±0.06	2.51±0.25
Spleen/body weight ratios(mg/100 g)	0.16±0.03	0.16±0.03	0.17±0.04	0.17±0.05	0.15±0.03
Kidney/body weight ratios(mg/100 g)	0.7±0.12	0.61±0.08	0.6±0.05	0.63±0.06	0.64±0.09
Uterus/body weight ratios(mg/100 g)	0.19±0.1	0.17±0.07	0.18±0.08	0.21±0.11	0.2±0.05
Ovary/body weight ratios(mg/100 g)	0.04±0.02	0.06±0.07	0.04±0.01	0.05±0.01	0.04±0.01

[a] Terminal body weight.

Supplementary Table S5 Summary of histopathology in kidney in the 90-day oral toxicity study (*n* = 10).

	Severity	Control	Cd_1	Cd_1+ Ca_L	Cd_1+ Ca_M	Cd_1+ Ca_H
Tubular vacuolation	±	10	10	9	7	7
	+	0	0	1	3	3

Protein cast	±	7	7	4	3	0
	+	3	3	6	7	10
Calcification	±	2	2	0	0	0
	+	8	6	9	5	4
	2+	0	2	1	3	4
	3+	0	0	0	2	2
Interstitial lymphocytic infiltrates	0	0	0	0	2	0

Severity scale: ±, Minimal; +, Slight; 2+, Moderate; 3+, Marked

Supplementary Table S6 Left femur dry weight in experiment 2 (g, n = 10).

Control	Cd_1	Cd_1+ Ca_M	Cd_5	Cd_5+ Ca_M	Cd_{50}	Cd_{50}+ Ca_M
0.63±0.06	0.69±0.07	0.66±0.04	0.66±0.07	0.68±0.08	0.62±0.04	0.62±0.07