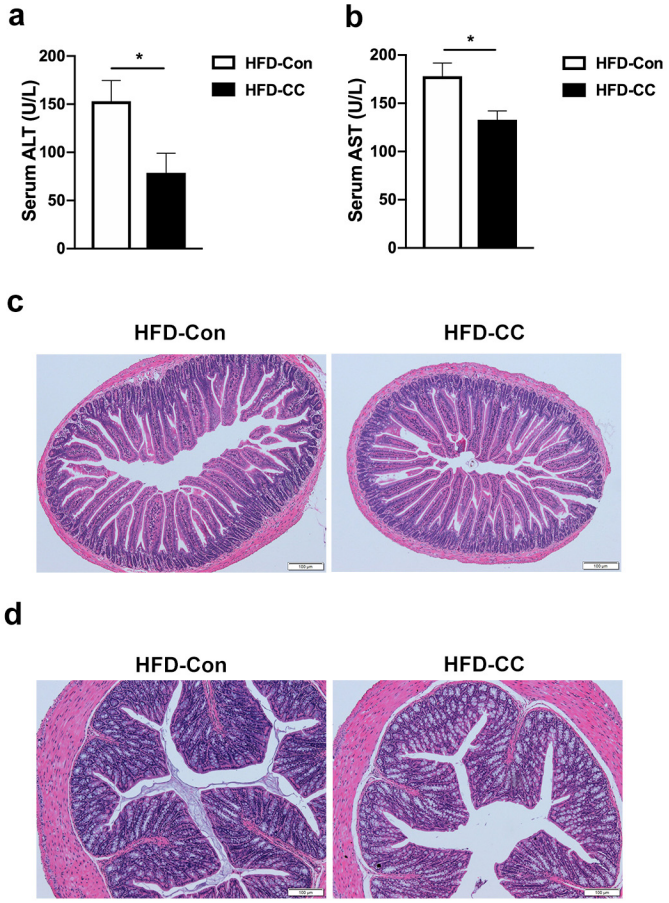
Electronic Supplementary Material (ESI) for Food & Function.

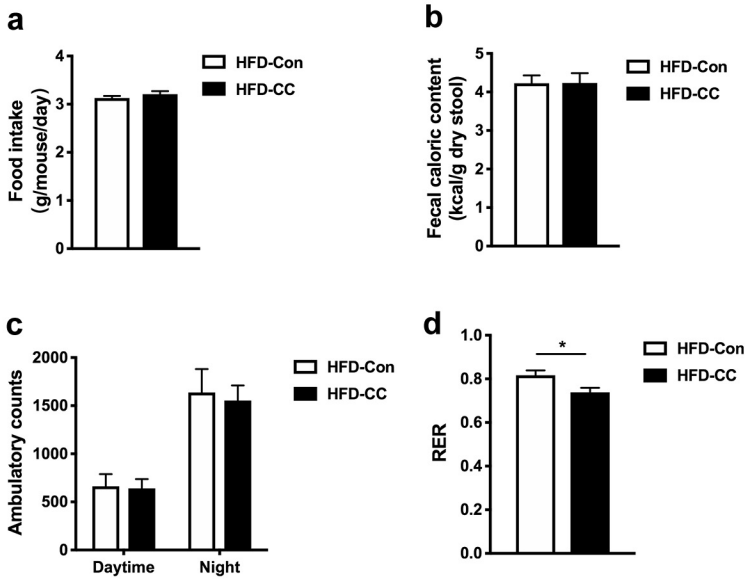
Supplementary Table 1: List of specific primers used for real-time quantitative PCR gene expression analysis, mtDNA copy number analysis and gut microbiota analysis.

Gene	Forward primer (5'-3')	Reverse primer (5'-3')
Ucp1	CTGCCAGGACAGTACCCAAG	TCAGCTGTTCAAAGCACACA
Ppargc1a	GGAGCTCCAAGACTCTAGACA	CCAAAGTCTCTCTCAGGTAGC
Prdm16	CAGCACGGTGAAGCCATTC	GCGTGCATCCGCTTGTG
Dio2	CAGTGTGGTGCACGTCTCCAATC	TGAACCAAAGTTGACCACCAG
Elovl3	TCCGCGTTCTCATGTAGGTCT	GGACCTGATGCAACCCTATGA
Cidea	ATCACAACTGGCCTGGTTACG	TACTACCCGGTGTCCATTTCT
Tjp1	GCTTTAGCGAACAGAAGGAGC	TTCATTTTTCCGAGACTTCACCA
Ocln	TTGAAAGTCCACCTCCTTACAGA	CCGGATAAAAAGAGTACGCTGG
Muc2	ATGCCCACCTCCTCAAAGAC	GTAGTTTCCGTTGGAACAGTGAA
Cyp7a1	GGGATTGCTGTGGTAGTGAGC	GGTATGGAATCAACCCGTTGTC
Cyp7b1	GGAGCCACGACCCTAGATG	GCCATGCCAAGATAAGGAAGC
Cyp8b1	CCTCTGGACAAGGGTTTTGTG	GCACCGTGAAGACATCCCC
Cyp27a1	GCACAGGAGAGTACGGAGG	CGGGCAAGTGCAGCACATA
Akr1d1	AAGACAGCTATTGATGAGGGGT	CCTCTTTACCTTCCCTTCTGCTA
Nrf1	GGTGTTTGGCGCAGCACCTT	CTCTGGGATAAATGCCCGAAGCT
Tfam	GCAAAGGATGATTCGGCTCAGG GAA	CCGGATCGTTTCACACTTCGACGG
Tfb2m	GTTCTTTGGCAAGTGGCCTG	ACTGATTCCCCGTGCTTTGA
Cd137	CGTGCAGAACTCCTGTGATAAC	GTCCACCTATGCTGGAGAAGG
Tmem26	ACCCTGTCATCCCACAGAG	TGTTTGGTGGAGTCCTAAGGTC
Rps18	TTCCAGCACATTTTGCGAGTA	CACGCCCTTAATGGCAGTGAT

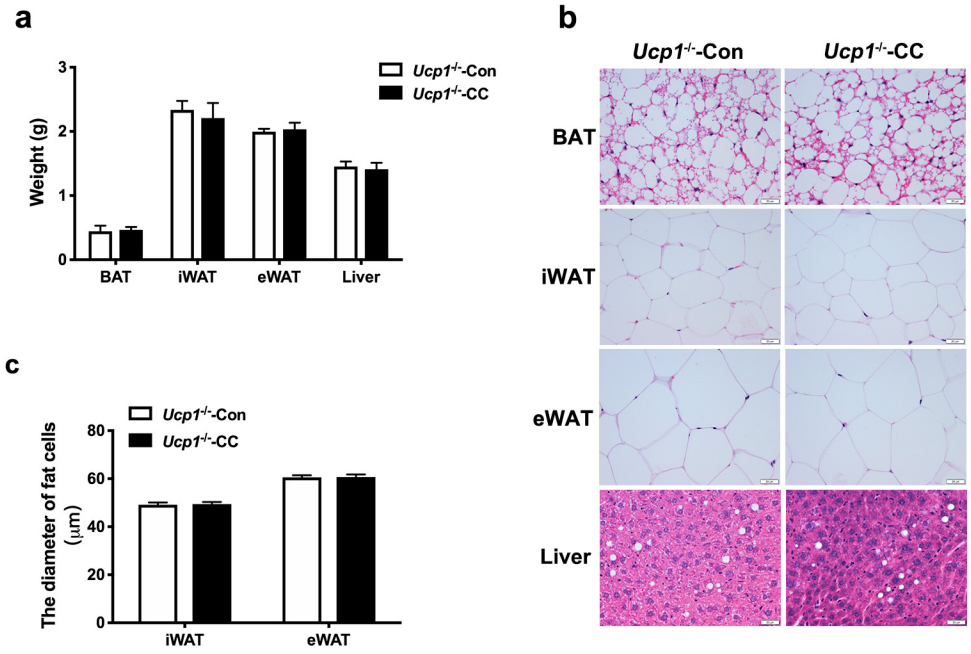
COX2 (mtDNA))	ATAACCGAGTCGTTCTGCCAAT	TTTCAGAGCATTGGCCATAGAA
Rps18 (mtDNA)	TGTGTTAGGGGACTGGTGGACA	CATCACCCACTTACCCCCAAAA
Verrucomicrobia	TCAKGTCAGTATGGCCCTTAT	CAGTTTTYAGGATTTCCTCCGCC
αProteobacteria	ACTCCTACGGGAGGCAGCAG	TCTACGRATTTCACCYCTAC
βProteobacteria	AACGCGAAAAACCTTACCTACC	TGCCCTTTCGTAGCAACTAGTG
δ,γProteobacteria	GCTAACGCATTAAGTRYCCCG	GCCATGCRGCACCTGTCT
Firmicutes	GCAGTAGGGAATCTTCCG	ATTACCGCGGCTGCTGG
Bacteroidetes	GTACTGAGACACGGACCA	ATTACCGCGGCTGCTGG
Actinobacteria	CGCGGCCTATCAGCTTGTTG	ATTACCGCGGCTGCTGG
Deferribacteres	CTATTTCCAGTTGCTAACGG	GAGHTGCTTCCCTCTGATTATG
Akkermansia	CAGCACGTGAAGGTGGGGAC	CCTTGCGGTTGGCTTCAGAT
Lactobacillus	AGCAGTAGGGAATCTTCCA	CACCGCTACACATGGAG
*Clostridium*_XIVa	AAATGACGGTACCTGACTAA	CTTTGAGTTTCATTCTTGCGAA
Bacteria 16S rDNA	ACTCCTACGGGAGGCAGCAG	ATTACCGCGGCTGCTGG



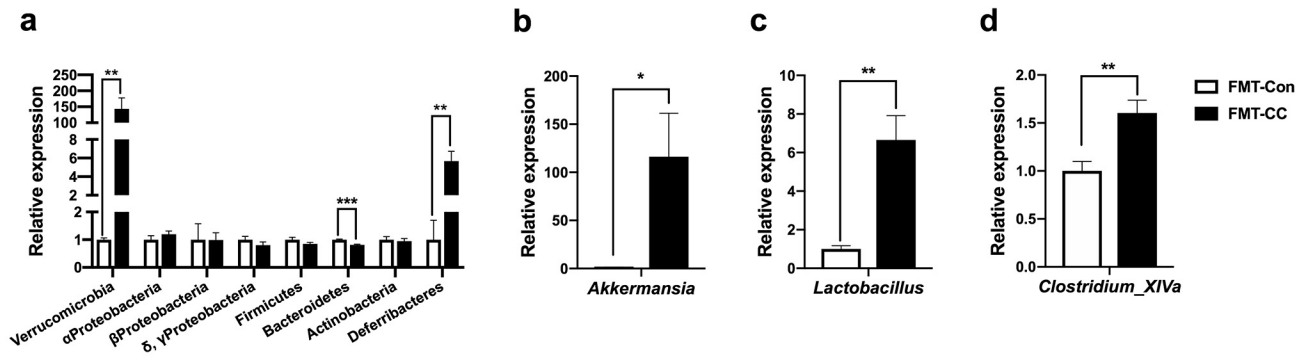
Supplementary Fig.1 Oral administration of curcumin does not lead to hepatotoxicity and intestinal toxicity. (a,b) Serum ALT (a) and AST (b) levels of HFD-fed mice treated with curcumin or vehicle for 12 weeks (n = 8/group). **(c,d)** Representative H&E staining of small intestine (c) and colon (d) from mice in (a,b). Scale bar, 100 μm. Numerical data are shown as the mean ± SEM. *$P < 0.05$.



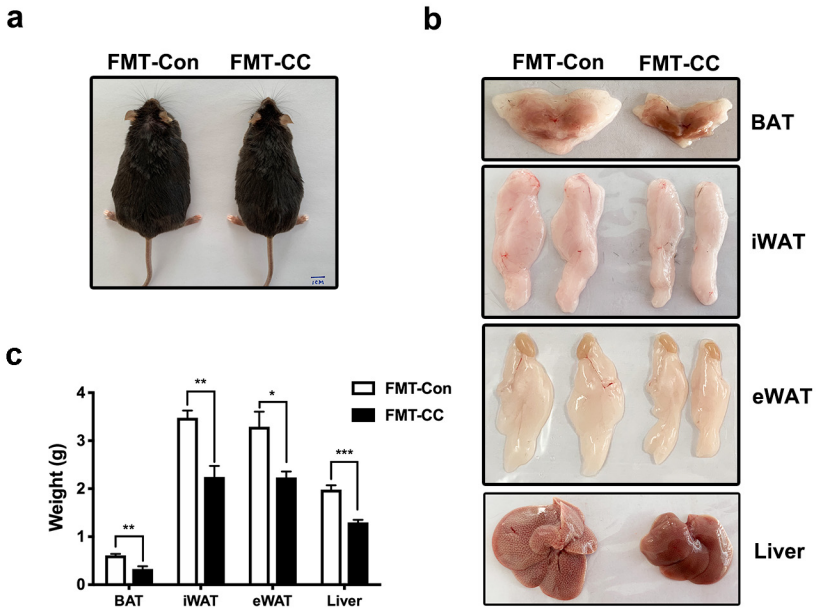
Supplementary Fig.2 Oral administration of curcumin does not affect food intake, fecal caloric content and total activity but reduces respiratory exchange ratio (RER) of HFD-fed mice. (a) Food intake of HFD-fed mice treated with curcumin or vehicle (n = 8/group). **(b)** Fecal caloric content of HFD-fed mice treated with curcumin or vehicle (n = 5/group). **(c)** Total activity of body weight-matched HFD-fed mice treated with curcumin or vehicle for 3 weeks (n = 8/group). **(d)** The average values of RER of mice in (c) (n = 8/group). Numerical data are shown as the mean ± SEM.*$P < 0.05$.



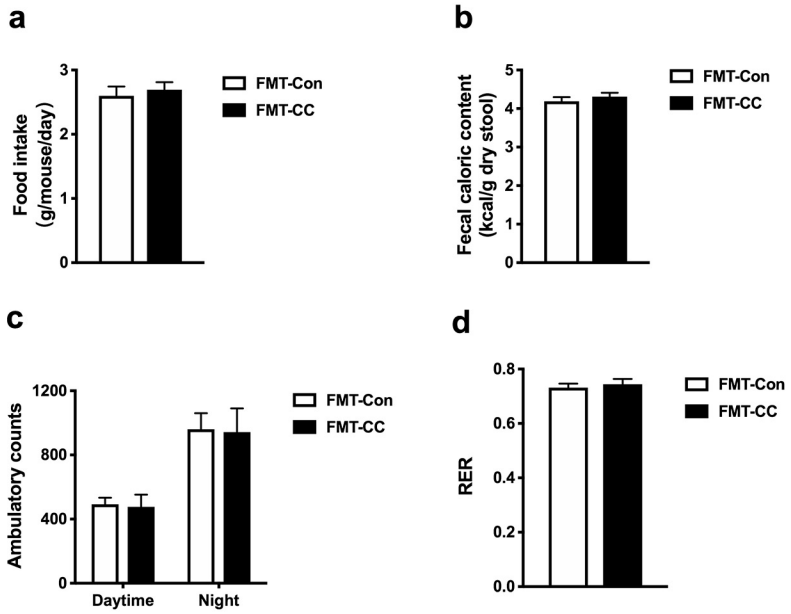
Supplementary Fig.3 *Ucp1* knockout removes the beneficial effects of oral administration of curcumin on HFD-induced obesity and hepatosteatosis. (a) The weight of BAT, iWAT, eWAT and liver from HFD-fed *Ucp1*$^{-/-}$ mice treated with curcumin or vehicle for 18 weeks (n = 6/group). **(b)** Representative H&E staining of BAT, iWAT, eWAT and liver from mice in (a). Scale bar, 20 μm. **(c)** Average diameters of fat cells from mice in (a). Numerical data are shown as the mean ± SEM.



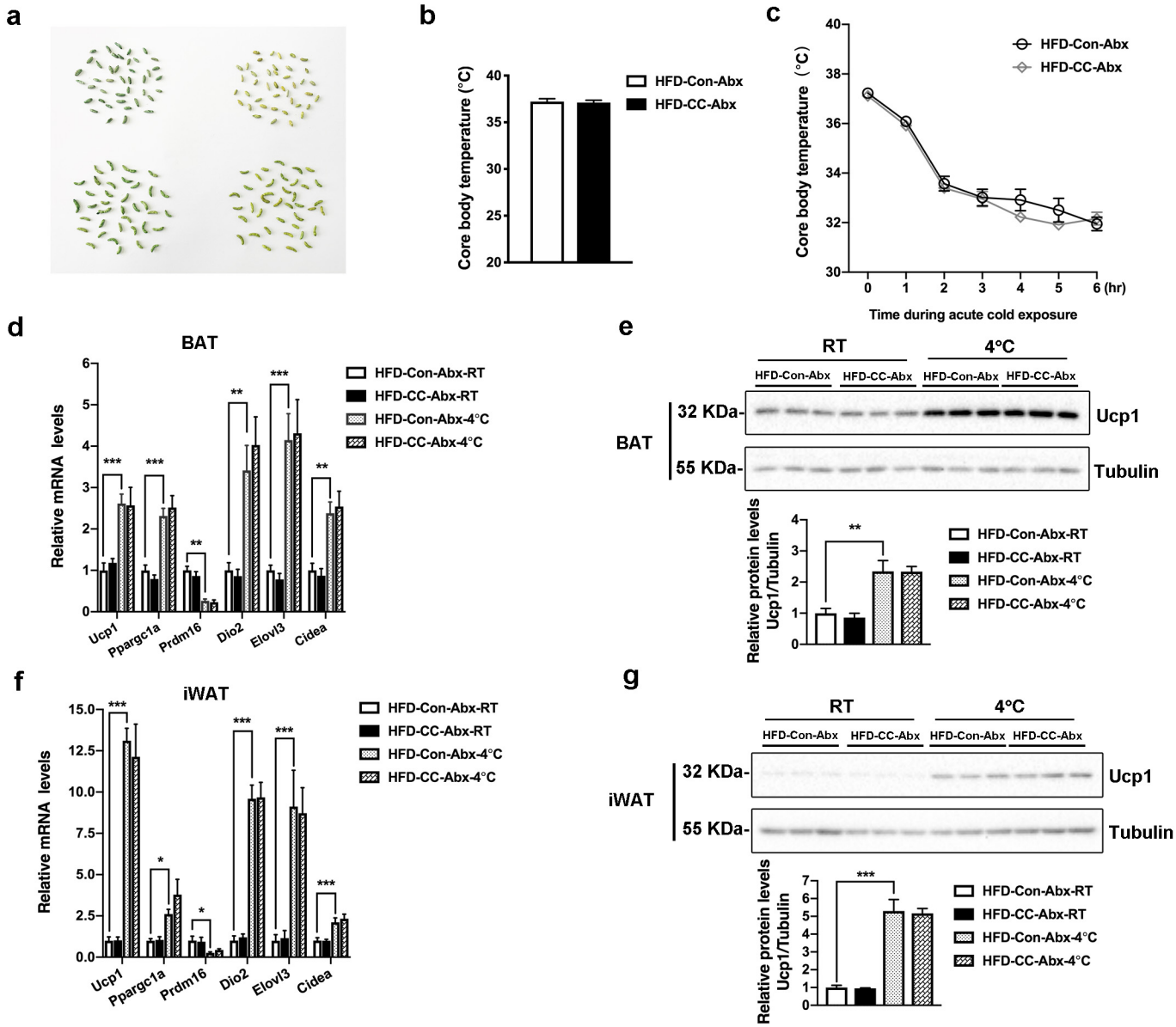
Supplementary Fig. 4 Transplantation with curcumin-shaped gut microbiota mimics the effects of oral administration of curcumin on gut microbiota. (a) Quantitative real-time PCR analysis of phylum-level modifications from 16S ribosomal DNA of fecal microbiome from endogenous gut microbiota-depleted mice after FMT experiments for 4 weeks (n = 6/group). **(b-d)** Quantitative real-time PCR analysis of genus *Akkermansia* (b), *Lactobacillus* (c) and *Clostridium*_XIVa (d) in fecal microbiome from mice in (a) (n = 6/group). Numerical data are shown as the mean ± SEM. **P* < 0.05, ***P* < 0.01, ****P* < 0.001.



Supplementary Fig. 5 Transplantation with curcumin-shaped gut microbiota ameliorates HFD-induced obesity and hepatosteatosis. (a) A representative photograph of endogenous gut microbiota-depleted HFD-fed mice colonized with microbiota harvested from curcumin- or vehicle-treated HFD-fed mice for 12 weeks. **(b)** Representative images of BAT, iWAT, eWAT with the testicles and liver from mice in (a). **(c)** The weight of BAT, iWAT, eWAT and liver of mice treated as in (a) (n = 6/group). Numerical data are shown as the mean ± SEM. $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.



Supplementary Fig.6 Transplantation with curcumin-shaped gut microbiota does not affect food intake, fecal caloric content, total activity and RER of endogenous gut microbiota-depleted HFD-fed mice. (a) Food intake of endogenous gut microbiota-depleted HFD-fed mice colonized with microbiota harvested from curcumin- and vehicle-treated HFD-fed mice (n = 8/group). **(b)** Fecal caloric content of mice treated as in (a) (n = 6/group). **(c)** Total activity of body weight-matched endogenous gut microbiota-depleted HFD-fed mice colonized with microbiota harvested from curcumin- and vehicle-treated HFD-fed mice for 3 weeks (n = 8/group). **(d)** The average values of RER of mice in (c) (n = 8/group). Numerical data are shown as the mean ± SEM.



Supplementary Fig.7 Antibiotics treatment eliminates the regulatory effects of oral administration of curcumin on Ucp1-dependent thermogenesis. **(a)** The morphology of the fecal pellet from vehicle (left panel) and curcumin-treated (right panel) HFD-fed mice before (upper panel) and after (lower panel) antibiotic treatment for 4 weeks. **(b)** The rectal temperature of vehicle- and curcumin-treated HFD-fed mice after antibiotic treatment (n = 8/group). **(c)** The rectal temperature of mice in (b) during acute cold exposure (n = 8/group). **(d)** Expression of thermogenic genes in BAT from mice treated as in (b) after acute cold exposure for 6 h (n = 6/group). **(e)** Western blotting (upper panel) and densitometric analyses (lower panel) of Ucp1 in BAT from mice in (d). **(f)** Expression of thermogenic genes in iWAT from mice treated as in (b) after acute cold exposure for 6 h (n = 6/group). **(g)** Western blotting (upper panel) and densitometric analyses (lower panel) of Ucp1 in iWAT from mice in (f). Numerical data are shown as the mean ± SEM. $*P < 0.05$, $**P < 0.01$, $***P < 0.001$.