

Oral glucose tolerance test, insulin tolerance test and the insulin resistance index

Blood samples were collected and the glucose was measured using a glucometer (Sannuo Biological Transmission Co., Ltd. Changsha, China). The oral glucose tolerance test: the mice were fasting for a whole night, and then orally administered glucose solution (2 g/kg body weight) at week 10. Their glucose was determined at 0, 15, 30, 60, 90 and 120 min after glucose administration. The insulin tolerance test: After fasting 6 h, the mice were intraperitoneally injected with insulin (0.5 U/kg body weight, Novolin R, Novo Nordisk) at week 11. Glucose was determined at 0, 15, 30, 60, 90 and, 120 min after insulin injection. The total glucose areas under the curve (AUC) were calculated using the trapezoidal rule. The insulin was tested using ELISA (Xin Le Biotechnology Co., Ltd. Shanghai, China).

HOMA-IR = (fasting insulin levels × fasting blood glucose)/22

Blood serum analysis

The serum levels of triglyceride (TG), total cholesterol (TC), LDL cholesterol (LDL-C), aspartate aminotransferase (AST), alanine aminotransferase (ALT), C-reactive protein (CRP) and creatinine (CRE) were determined using Hitachi-7180 automatic biochemical analyzer (Tokyo, Japan). The serum levels of adiponectin, LPS, interleukin-1 β (IL-1 β), interleukin-6 (IL-6), tumor necrosis factor-alpha (TNF- α) were tested using ELISA (Xin Le Biotechnology Co., Ltd. Shanghai, China).

Morphology of the liver and epididymal fat

After mice were sacrificed, the liver and epididymal fat were subsequently fixed in 4% paraformaldehyde overnight at 4°C, and then embedded in paraffin for hematoxylin and eosin (H&E) staining analysis. The stained area was viewed and photographed using a microscope (Olympus Corporation, Tokyo, Japan) under the objective.

Quantitative PCR analysis

Total RNA was extracted from liver and epididymal fat using RNAiso Plus reagent (TaKaRa, Dalian, China). Two μ L RNA after an evaluation of quality determined using a Micro-Spectrophotometer Nano-200 was used to synthesize cDNA with the Fast King RT Kit (With gDNase, TIANGEN, Beijing, China). Two μ L of the diluted cDNA (1:10) was done quantitative PCR using SYBR Green BioEasy Master mix (BIOER, Hangzhou, China) and a CFX96 TouchTM Real-Time PCR Detection System (Bio-Rad). The sequences of the primers used for Real Time qPCR of this study are shown in Table S2. The level of mRNA expression was normalized with housekeeping gene β -actin, and data analysis was using the 2- $\Delta\Delta Ct$ method.

Short-chain fatty acid concentration of feces

Two hundred mg fecal samples were homogenized with 2 mL distilled water and centrifuged at 4°C at 10000 $\times g$ for 10 min. Taken 1 mL supernatants and acidified by adding 200 μ L 50% H₂SO₄ (v/v). After vortexing and standing for 5 min, 1 mL diethyl ether was added, and the mixed solution was incubated at 4°C for 30 min, and then the samples were centrifuged at 4°C (10000 $\times g$, 10 min). The organic phase was filtered using a 0.22 μ m nylon filter and collected for gas chromatography analysis (GC-2014C, Shimadzu Corporation, Japan; 30 m \times 0.25 μ m \times 0.25 μ m RTX-wax column; flame ionization detector). The initial temperature was 50°C for 1 min, then increased 120°C at 15°C/min, 170°C at 5°C/min, 240°C at 15°C/min, and then held at 3 min. The injector temperature was 250°C and the detector temperature was 270°C.

Table S1 The chemical sensors corresponding to different types of volatile substances in electronic nose

Sensor name	Sensor sensitivities
W1C	Sensitive to aromatic organic compounds
W5S	Sensitive to nitrogen oxides
W3C	Sensitive to ammonia and aromatic compounds
W6S	Sensitive to hydrogen
W5C	Sensitive to alkanes, aromatic compounds, and nonpolar organic compounds
W1S	Sensitive to methane
W1W	Sensitive to sulfides and terpenes
W2S	Sensitive to alcohol and partially sensitive to aromatic compounds
W2W	Sensitive to aromatic compounds and organic sulfides
W3S	Sensitive to alkanes

Table S2. Sequences of the primers used for quantitative Real-Time PCR

Target gene	Forward primer (5'→3')	Revere primer (5'→3')	Product Size (bp)	Accession number
TNF-α	CCCTCACACTCAGATCATCTTCT	GCTACGACGTGGGCTACAG	61	NM_013693.3
IL-1β	CCTGCAGCTGGAGAGTGTGGAT	TGCTCTGCTTGTGAGGTGCTG	150	NM_008361.4
IL-6	GGCCTTCCCTACTTCACAAG	ATTTCCACGATTCCCAGAG	126	NM_001314054.1
FAS	GGAGGTGGTGATAGCCGGTAT	TGGGTAATCCATAGAGCCCAG	140	NM_007988.3
Fabp2	TTCTCAGAGCCTGGAGCAAC	GATGACGAATGAGCCTGGCA	86	NM_007980.3
CD36	CAGATGACGTGGCAAAGAAC	TGGCTCCATTGGGCTGTA	144	NM_001159558.1
PPARγ	CGCTGATGCACTGCCTATG	ATGCGAGTGGCTTCCATCA	124	NM_011146.3
MCP-1	CAGGTCCCTGTATGCTTCT	CCCATTCTTCTGGGGTCA	121	NM_011333.3
Srebp-1c	GATCAAAGAGGAGGCCAGTGC	TAGATGGTGGCTGCTGAGTG	191	NM_001358315.1
β-actin	GGACTGTTACTGAGCTGCGTT	CGCCTCACCGTTCCAGTT	209	NM_007393.5

Table S3. OAV analysis of aroma characteristics of different teas

Compounds	Odor detection thresholds		OAV	
			FBT	MFBT
Acetic acid	180		0.0131	-
Tetradecanoic acid	10.0		0.0653	-
n-Hexadecanoic acid	>10		-	<0.4294
Benzene	2		-	0.1160
Toluene	1		0.6720	0.4340
o-Xylene	0.45023		1.9546	-
Naphthalene, 1-methyl-	0.02		63.6000	-
Naphthalene, 2-methyl-	0.01		-	27.9000
Hexanoic acid, methyl ester	0.087		-	2.6782
methyl salicylate	40		-	0.0165
2-Heptanone	10		0.3958	0.1708
5-Hepten-2-one, 6-methyl-	0.05		66.1400	81.0200
2-Octanone	0.0145		-	16.6897
Cyclohexanone, 2,2,6-trimethyl-	0.1		-	19.7300
Isophorone	11.9		-	0.0715
3-Buten-2-one, 4-(2,6,6-trimethyl-1-cyclohexen-1-yl)-	0.0084		-	156.7857
Propanal, 2-methyl-	0.018		-	41.1667
Butanal, 3-methyl-	1.1		-	4.0136
Butanal, 2-methyl-	0.0125		-	396.1600
Pentanal	0.012		-	50.0833
Hexanal	5		1.7144	1.8476
Benzaldehyde	0.33		5.3394	10.0364
Benzeneacetaldehyde	0.06		-	41.1833
2-Octenal, (E)-	0.003		223.3333	-
Nonanal	0.02		418.9500	-
Decanal	0.03		86.0667	13.2333
1-Cyclohexene-1-carboxaldehyde, 2,6,6-trimethyl-	0.003		-	396.3333
1,3-Butanediol	10-20		0.0234-0.1172	-
1-Octanol	0.13		7.5231	2.7846
trans-Linalool oxide (furanoid)	0.32		13.0750	7.5844
Linalool	6		-	0.6598
Furan, 2-methyl-	27			0.0274
Furan, 2-ethyl-	8		0.1264	0.3663
Furan, 2-pentyl-	0.1		16.5100	25.4500
D-Limonene	10		-	0.0467

Table S4 Go groups and KEGG pathways of the fecal microbial community

	Name	C †	M	FBT	MFBT
Go terms	General function prediction only	0.11371046 7	0.11609896 4	0.11850390 6	0.11725424 5
	Carbohydrate transport and metabolism	0.08421690 9	0.08139034 9	0.08784544 3	0.09021665 6
	Cell wall/membrane/envelope biogenesis	0.07821097 8	0.06958315	0.06778546 6	0.06850963
	Amino acid transport and metabolism	0.07541812 8	0.07937497 4	0.07841126 4	0.07721695 9
	Transcription	0.07288561 9	0.07772000 7	0.08265281 3	0.08336249 2
	Translation, ribosomal structure and biogenesis	0.07140408 8	0.06606493	0.06758351 8	0.06613225 9
	Replication, recombination and repair	0.06706729 4	0.06630560 9	0.06929896 6	0.06758530 1
	Function unknown	0.06425626 4	0.06673518 6	0.068486	0.06885251
	Energy production and conversion	0.05811516	0.05668554 7	0.05241207 3	0.05254662 7
	Signal transduction mechanisms	0.05052676 4	0.06342567 6	0.05778577 1	0.05750799 8
	Inorganic ion transport and metabolism	0.04629827 3	0.04377824 1	0.04209692	0.04380659 4
	Coenzyme transport and metabolism	0.04345909 2	0.04197970 7	0.03861576 6	0.03913292 3
	Posttranslational modification, protein turnover, chaperones	0.03432732 4	0.03201289 2	0.03095025 9	0.03113896
	Nucleotide transport and metabolism	0.03130933 2	0.02914904 5	0.03119605 7	0.03021078 7
	Defense mechanisms	0.02942144 3	0.02943872 1	0.03259356 4	0.03160797 6
	Lipid transport and metabolism	0.02402233 7	0.02136839 9	0.02096908 8	0.02171522 6
	Intracellular trafficking, secretion, and vesicular transport	0.02165763 1	0.01939259 3	0.01753166 5	0.01837476 5
	Cell cycle control, cell division, chromosome partitioning	0.01323491 9	0.01328235 6	0.01382443 7	0.01329960 3
	Cell motility	0.01093704 9	0.01585388 1	0.01215462 4	0.01168103 9

	Secondary metabolites biosynthesis, transport and catabolism	0.0092763	0.01003862 8	0.00905931 9	0.00955261 5
	Chromatin structure and dynamics	9.08E-05	0.00019759 8	0.00010582 1	0.00011637 9
	Cytoskeleton	7.90E-05	7.10E-05	8.75E-05	0.00010248 8
	RNA processing and modification	7.48E-05	5.26E-05	4.97E-05	7.59E-05
	Extracellular structures	1.63E-08	1.79E-08	5.65E-08	6.74E-08
	Carbohydrate metabolism	0.150377	0.15177	0.157306	0.159428
	Global and overview maps	0.137685	0.135496	0.134312	0.135548
	Amino acid metabolism	0.110041	0.10812	0.10853	0.108848
	Energy metabolism	0.072048	0.0701	0.069133	0.069338
	Metabolism of cofactors and vitamins	0.06606	0.064904	0.061181	0.061449
	Nucleotide metabolism	0.060034	0.057624	0.059945	0.058854
	Translation	0.056855	0.056007	0.057903	0.056339
	Replication and repair	0.048466	0.046363	0.049156	0.048088
	Membrane transport	0.040192	0.052548	0.055165	0.052718
	Lipid metabolism	0.032912	0.030964	0.031176	0.032458
	Glycan biosynthesis and metabolism	0.032874	0.024979	0.024672	0.025695
	Signal transduction	0.027462	0.033116	0.028668	0.028576
	Folding, sorting and degradation	0.022027	0.021419	0.021276	0.021307
	Metabolism of other amino acids	0.021787	0.020542	0.021319	0.021387
	Metabolism of terpenoids and polyketides	0.018733	0.018362	0.019261	0.018892
	Xenobiotics biodegradation and metabolism	0.015636	0.017327	0.016741	0.017426
	Biosynthesis of other secondary metabolites	0.014312	0.013377	0.013398	0.013608
	Cell motility	0.012535	0.021004	0.015552	0.01429
	Drug resistance	0.008761	0.007556	0.007608	0.00763
	Infectious diseases: Bacterial	0.007957	0.008284	0.00797	0.007599
	Cancers: Overview	0.007822	0.007408	0.007775	0.007892
	Cell growth and death	0.007743	0.00749	0.00731	0.007223
	Transport and catabolism	0.006946	0.004547	0.004374	0.00484
	Endocrine system	0.004884	0.004855	0.004673	0.004778

Nervous system	0.003344	0.003573	0.003716	0.003691
Neurodegenerative diseases	0.002423	0.002052	0.001852	0.001981
Transcription	0.002373	0.002466	0.002587	0.002501
Endocrine and metabolic diseases	0.00185	0.001553	0.001605	0.00163
Environmental adaptation	0.001818	0.002192	0.001991	0.001933
Immune system	0.001339	0.001296	0.001355	0.00134
Cancers: Specific types	0.000782	0.000964	0.001008	0.000926
Immune diseases	0.000659	0.000598	0.000649	0.000624
Digestive system	0.000504	0.000366	0.000257	0.000297
Excretory system	0.000436	0.000222	0.000225	0.000323
Infectious diseases:				
Parasitic	0.000166	0.000152	0.000142	0.000218
Circulatory system	0.000107	7.33E-05	6.21E-05	7.26E-05
Substance dependence	3.65E-05	0.000318	0.000136	0.000238
Infectious diseases: Viral	1.31E-05	1.37E-05	1.24E-05	1.34E-05

[†] C, mice fed with standard diet; M, mice fed with high-fat diet and 10% fructose in drinking water (HFFD); FBT, mice fed with HFFD and FBT; MFBT, mice fed with HFFD and MFBT.

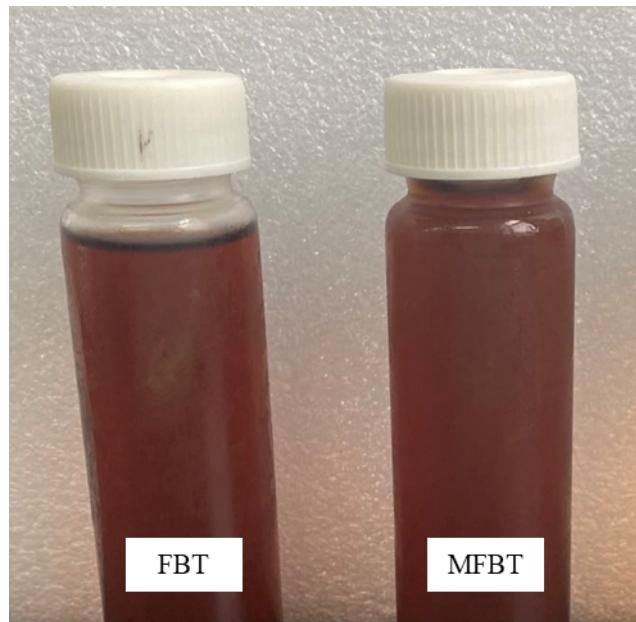


Fig. S1. The color of the FBT and MFBT.

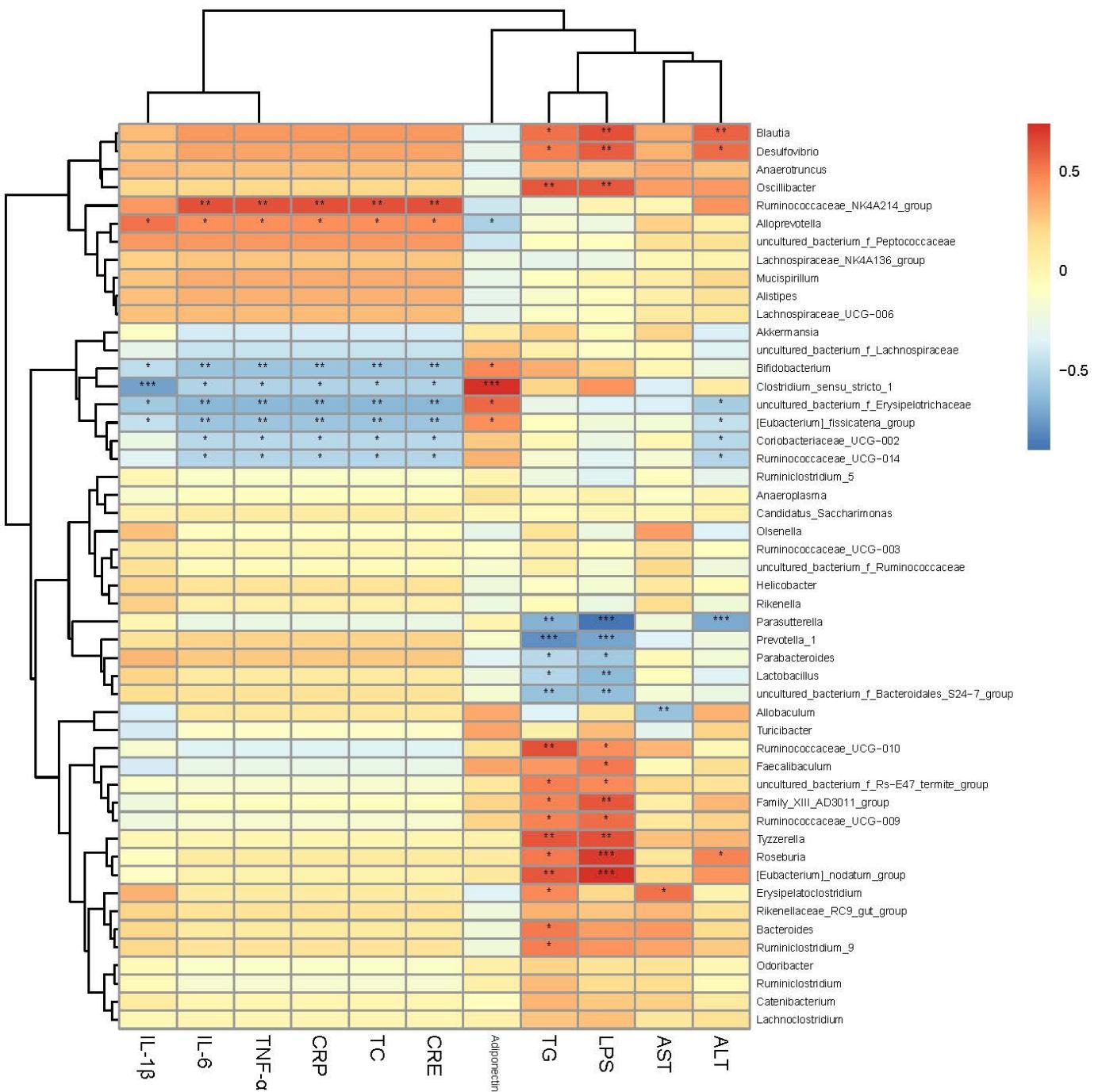


Fig. S2. Correlation analysis between obesity-related parameters and gut microbiota at the genus level. The color ranges from blue to red, indicating the change in correlation from negative to positive. *, **, and *** represent $0.01 < P \leq 0.05$, $0.001 < P \leq 0.01$, and $P \leq 0.001$, respectively.