

Mulberry leaf alleviates streptozotocin-induced diabetic rats by attenuating NEFA signaling and modulating intestinal microflora

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Running Title: Mulberry leaf alleviates diabetes by reducing NEFAs and restoring intestinal microbiota communities

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Table S1. Methods for Composition Analysis

Composition parameters	Method
Moisture	Chinese standard GB 5009.3-2010. 2010. National food safety standard: Determination of moisture in foods. Standards Press of China, Beijing, China.
Ash	Chinese standard GB 5009.4-2010. 2010. National food safety standard: Determination of ash in foods. Standards Press of China, Beijing, China.
Protein	Chinese standard GB 5009.5-2010. 2010. National food safety standard: Determination of protein in foods. Standards Press of China, Beijing, China.
Crude Fiber	Chinese standard GB/T 5009.10-2003. 2003. National food safety standard: Determination of crude fiber in vegetable foods. Standards Press of China, Beijing, China.
Fat	Chinese standard GB/T 5009.6-2003. 2003. National food safety standard: Determination of fat in foods. Standards Press of China, Beijing, China.
Calcium	Chinese standard GB/T 5009.92-2003. 2003. National food safety standard: Determination of calcium in foods. Standards Press of China, Beijing, China.
Zinc	Chinese standard GB/T 5009.91-2003. 2003. National food safety standard: Determination of zinc in foods. Standards Press of China, Beijing, China.
Copper	Chinese standard GB/T 5009.13-2003. 2003. National food safety standard: Determination of copper in foods. Standards Press of China, Beijing, China.
Manganese	Chinese standard GB/T 5009.90-2003. 2003. National food safety standard: Determination of iron, magnesium and manganese in foods. Standards Press of China, Beijing, China.
Potassium	Chinese standard GB/T 5009.91-2003. 2003. National food safety standard: Determination of potassium and sodium in foods. Standards Press of China, Beijing, China.
Phosphorus	Chinese standard GB/T 5009.87-2003. 2003. National food safety standard: Determination of phosphorus in foods. Standards Press of China, Beijing, China.
Vitamin E	Chinese standard GB/T 5009.82-2003. 2003. National food safety standard: Determination of retinol and tocopherol in foods. Standards Press of China, Beijing, China.
Vitamin B1	Chinese standard GB 5413.11-2010. 2010. National food safety standard: Determination of vitamin B1 in foods for infants and young children, milk and milk products. Standards Press of China, Beijing, China.
Vitamin B2	Chinese standard GB 5413.12-2010. 2010. National food safety standard: Determination of vitamin B2 in foods for infants and young children, milk and milk products. Standards Press of China, Beijing, China.
Fatty acids	Chinese standard GB/T 21514-2008. 2008. Determination of fatty acids in feeds. Standards Press of China, Beijing, China.
Polysaccharides	Chinese standard NY/T 1676-2008. 2008. National food safety standard:

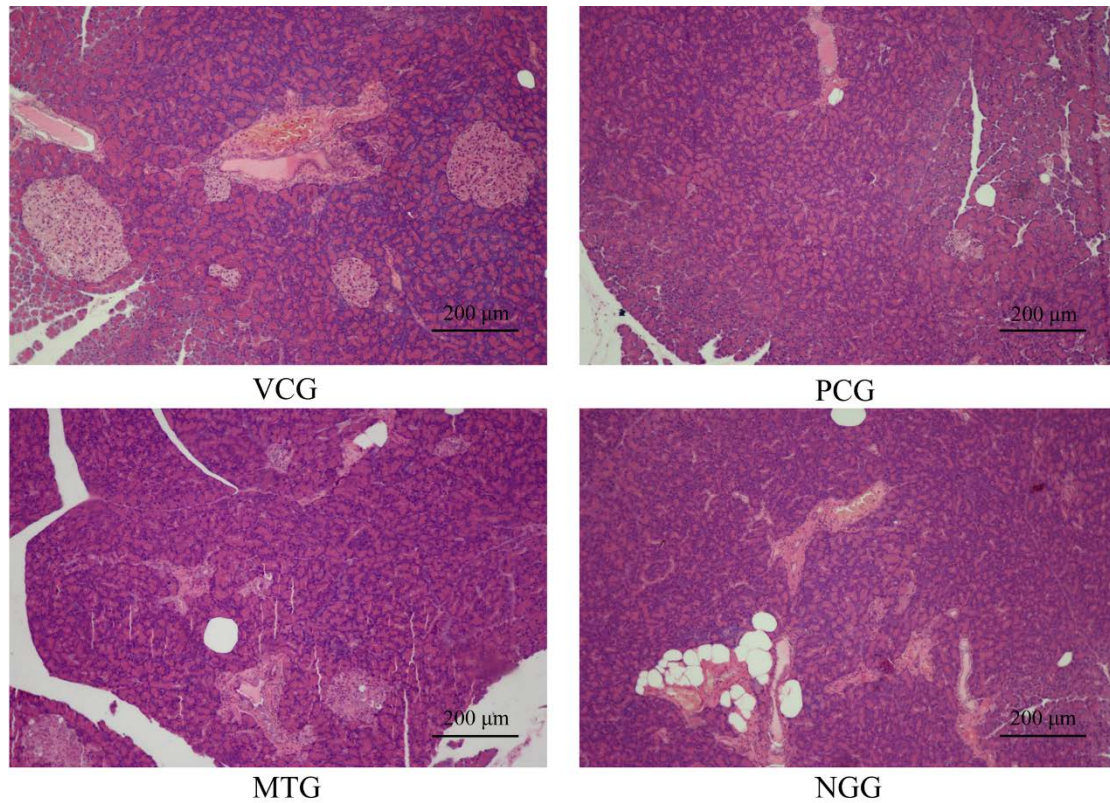


Figure S1. Histological data on pancreas (H & E stain). Diabetic rat induced by STZ posed significantly damaged pancreas islet as showed in figure S1 (NCG, PCG and MTG). The number and size of pancreas islet were decreased; mass of necrocytosis were observed in pancreas islet β -cell; in addition, vacuolization was found in pancreas in NCG. However, mulberry leaf treatment could inhibit necrocytosis and vacuolization indicating beneficial effect in protecting pancreas and pancreas islet damaged by STZ.

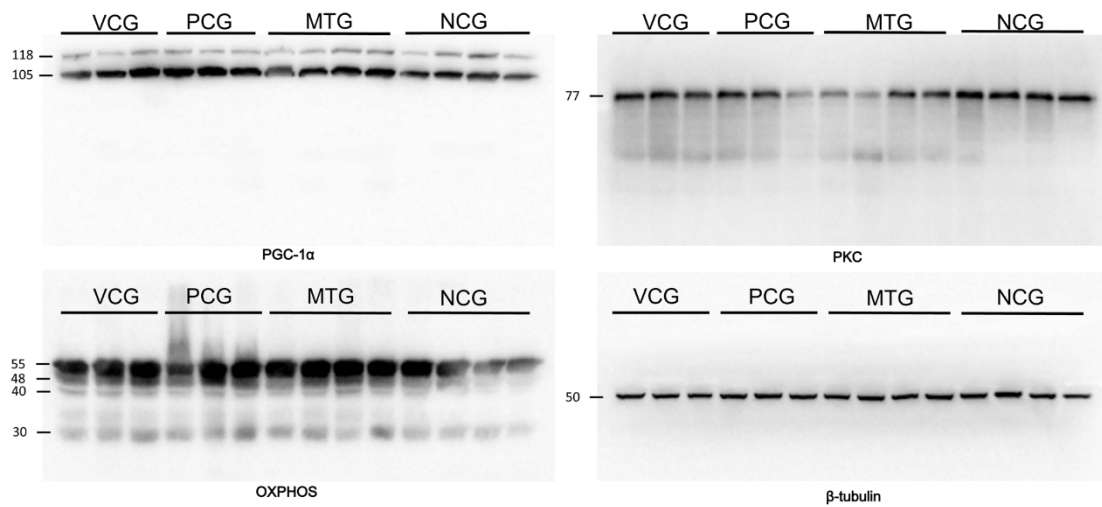


Figure S2. Western blot for protein in liver tissue, mulberry leaf decreased PKC and increased OXPPOS expression. β -tubulin serves as a loading control. For presentation purposes additional lanes were excised. PKC, PGC-1 α , OXPPOS and β -tubulin were cropped from different parts of the same gel. These proteins were stained one by one after using Restore™ Western Blot Stripping Buffer (Thermo Scientific, USA).

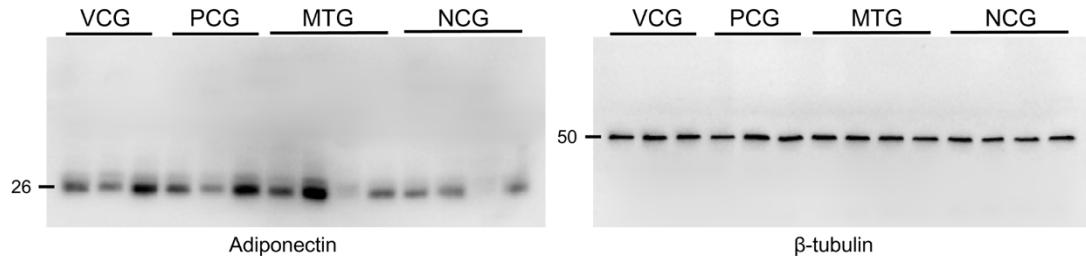


Figure S3. Western blot for protein in subcutaneous fat, mulberry leaf increased adiponectin expression in subcutaneous fat. β -tubulin serves as a loading control. For presentation purposes additional lanes were excised. Adiponectin and β -tubulin were cropped from different parts of the same gel. Both proteins were stained one by one after using Restore™ Western Blot Stripping Buffer (Thermo Scientific, USA).

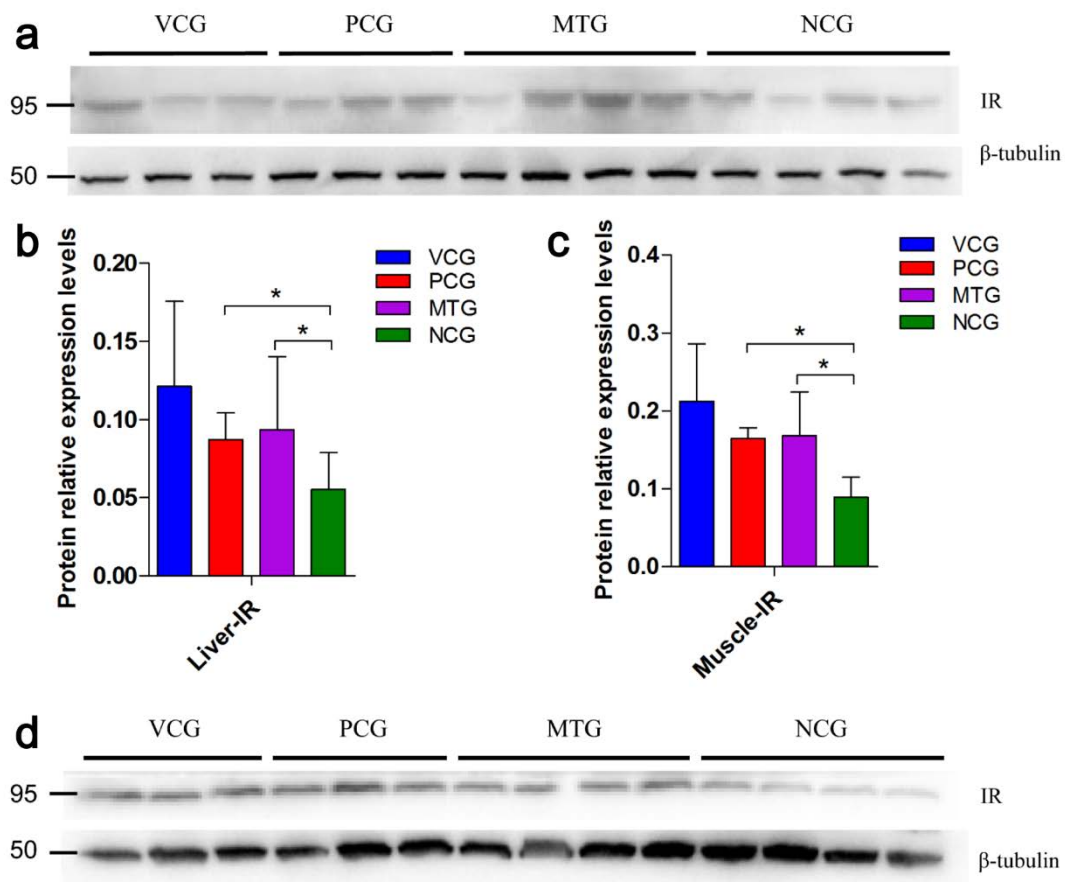


Figure S4. Insulin receptors (IR) expression in peripheral tissue. **(a, b)** Western blot for insulin receptor (IR) in liver tissue. **(c, d)** Western blot for insulin receptor (IR) in muscle tissue. IR was significantly ($P < 0.05$) up-regulated in peripheral tissue (liver and muscle) of both mulberry leaf and glibenclamide treatments groups at post-translation level. β -tubulin serves as a loading control. For presentation purposes additional lanes were excised. IR and β -tubulin were cropped from different parts of the same gel. Both proteins were stained one by one after using Restore™ Western Blot Stripping Buffer (Thermo Scientific, USA). * $P < 0.05$ compared with NCG. Value=mean \pm SD (N=3-4).

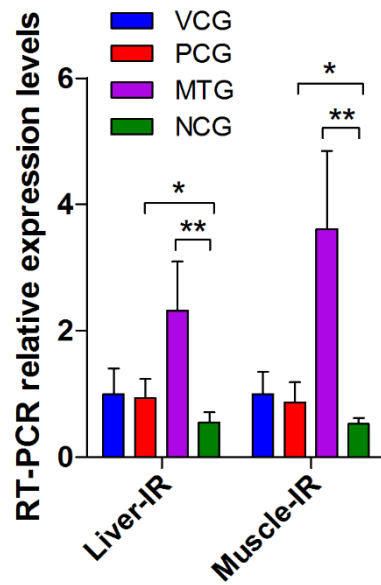


Figure S5, IR expression on RT-PCR, IR was significantly up-regulated in peripheral tissue (liver and muscle) of both mulberry leaf ($P < 0.01$) and glibenclamide ($P < 0.05$) treatments groups at transcription level. * $P < 0.05$ compared with NCG, ** $P < 0.01$ compared with NCG. Value=mean \pm SD (N=6).

Table S2. The primers used in reverse transcription-PCR analysis

Gene	Primer sequences (forward)	Primer sequences (Reverse)	Amplicon length (bp)	TM ($^{\circ}$ C)
Adiponectin	CTACTGTTGCAAGCTCTCC	CTTCACATCTTTCATGTACACC	513	55
ADPR1	CTTCTACTGCTCCCCACAGC	TCCAGGAACACTCCTGCTC	139	60
ADPR2	TGTAAGGTGTGGGAAGGTCG	GGAAAGAAGGCATAGGAGGC	112	58
PGC-1 α	AGTTTTTGGTGAAATTGAGGAAT	TCATACTTGCTCTTGGTGGAAGC	249	58
AK2	GTGACCTTGCTGTCTGAGAA	TCCGCTTGTGGAATACTACA	240	57
PKC1	CAACCAAGAGGAGGGTGAAT	CACTGTCTGGAAGCAGGAGT	397	58
IR	GGCGAGAAGACCATTGATT	AAGTGACACCAGAGCATAGGA	189	58
β -actin	GTCAGGTCATCACTATCGGCAAT	AGAGGTCTTTACGGATGTCAACGT	147	60

Table S3. Nutritional proximates, vitamins, minerals of mulberry leaf powder. Value=mean \pm SD (N=3)

Substance	Concentration
Protein (%)	19.6 \pm 1.6
Crude fibre (%)	11.200 \pm 0.889
Lipid (%)	1.780 \pm 0.153
Vitamin b1 (mg/100g)	0.002 \pm 0
Vitamin b2 (mg/100g)	0.738 \pm 0.082
Moisture (%)	9.460 \pm 0.681
Ash (%)	9.480 \pm 0.549
Kalium (mg/kg)	12060 \pm 1086
Calcium (mg/kg)	16800 \pm 8075
Sodium (mg/kg)	133.8 \pm 9.81
Magnesium (mg/kg)	5201 \pm 386
Ferrum (mg/kg)	316.7 \pm 27.5
Manganese (mg/kg)	44.5 \pm 2.86
Zinc (mg/kg)	30.6 \pm 1.46
Cuprum (mg/kg)	9.6 \pm 0.56
Phosphorus (mg/kg)	4138 \pm 126

Table S4. Fatty acids of mulberry leaf powder. Value=mean \pm SD (N=3)

Fatty acids	Percentage (%)
C14:0	0.46 \pm 0.021
C16:0	18.12 \pm 1.26
C16:1	0.12 \pm 0.007
C17:0	0.28 \pm 0.011
C18:0	3.69 \pm 0.165
C18:1	3.00 \pm 0.196
C18:2	23.00 \pm 1.83
C18:3	39.31 \pm 2.06
C20:0	1.54 \pm 0.061
C22:0	1.16 \pm 0.087
Others	9.32 \pm 0.646

Table S5. Body weight (g) changes of all animals. Value=mean \pm SD (N=6)

weeks	0	1	2	3	4	5	6	7	8	9	10	11	12	13
VCG	155 \pm 12	219 \pm 15	285 \pm 22	340 \pm 26	364 \pm 26	375 \pm 28	398 \pm 31 ^a	419 \pm 33 ^a	440 \pm 41 ^a	463 \pm 44 ^a	483 \pm 46 ^a	498 \pm 47 ^a	512 \pm 45 ^a	523 \pm 51 ^a
PCG	155 \pm 9	197 \pm 10	256 \pm 18	319 \pm 22	359 \pm 25	338 \pm 34	342 \pm 58 ^{ab}	360 \pm 48 ^{ab}	377 \pm 52 ^{ab}	383 \pm 55 ^{ab}	387 \pm 64 ^b	395 \pm 68 ^b	406 \pm 65 ^b	412 \pm 70 ^b
MTG	158 \pm 11	203 \pm 16	262 \pm 22	326 \pm 27	367 \pm 25	333 \pm 38	320 \pm 33 ^b	373 \pm 42 ^{ab}	383 \pm 55 ^{ab}	398 \pm 58 ^{ab}	406 \pm 64 ^{ab}	411 \pm 65 ^b	419 \pm 68 ^b	425 \pm 73 ^b
NCG	151 \pm 10	197 \pm 14	252 \pm 20	317 \pm 25	360 \pm 25	324 \pm 26	300 \pm 35 ^b	263 \pm 36 ^b	321 \pm 44 ^b	342 \pm 48 ^b	348 \pm 40 ^b	371 \pm 42 ^b	376 \pm 45 ^b	381 \pm 47 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S6. Glucose tolerance test (GTT, mmol/L) of all animals. Value=mean \pm SD (N=6)

Time (min)	0	15	30	60	90	120	Area under the curve
VCG	4.2 \pm 0.43 ^a	14.9 \pm 1.58 ^a	10.7 \pm 1.92 ^a	6.5 \pm 1.17 ^a	5.43 \pm 0.63 ^a	5.22 \pm 0.39 ^a	1091 \pm 117 ^a
PCG	9.7 \pm 3.79 ^{ab}	22.7 \pm 4.22 ^{ab}	21.2 \pm 3.9 ^{ab}	19.9 \pm 3.55 ^{ab}	18.5 \pm 3.51 ^{ab}	16.22 \pm 3.7 ^{ab}	2808 \pm 311 ^{ab}
MTG	10.1 \pm 3.62 ^{ab}	21.8 \pm 2.53 ^{ab}	24.1 \pm 4.46 ^b	21.6 \pm 4.83 ^{ab}	20.23 \pm 3.97 ^{ab}	16.7 \pm 3.01 ^{ab}	3002 \pm 467 ^{ab}
NCG	16.8 \pm 1.2 ^b	26.6 \pm 2.12 ^b	27.4 \pm 0.86 ^b	27.2 \pm 3.07 ^b	26.18 \pm 1.71 ^b	23.98 \pm 1.48 ^b	3854 \pm 144 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S7. Fasting blood glucose levels (mmol/L) of all animals. Value=mean \pm SD (N=6)

Time (day)	F	0	3	14	30	45	60
VCG	4.7 \pm 0.38	5.18 \pm 0.25 ^a	5.23 \pm 0.66 ^a	5.05 \pm 0.37 ^a	5.4 \pm 0.48 ^a	4.75 \pm 0.5 ^a	4.56 \pm 0.52 ^a
PCG	5.19 \pm 0.43	14.5 \pm 6.27 ^b	17.14 \pm 5.89 ^b	16.91 \pm 5.94 ^b	14.63 \pm 5.64 ^{ab}	11.24 \pm 5.7 ^{ab}	9.94 \pm 4.83 ^{ab}
MTG	5.1 \pm 0.46	14.44 \pm 4.43 ^b	15.21 \pm 4.47 ^b	14.99 \pm 5.06 ^b	12.74 \pm 3.68 ^{ab}	10.83 \pm 4.3 ^{ab}	10.08 \pm 3.23 ^{ab}
NCG	4.76 \pm 0.48	14.48 \pm 6.22 ^b	21.81 \pm 7.18 ^b	19.3 \pm 5.66 ^b	20.4 \pm 1.63 ^b	19.94 \pm 1.38 ^b	18.64 \pm 2.43 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S8. HbA1c proportion (%) of all animals. Value=mean \pm SD (N=6)

	VCG	PCG	MTG	NCG
sample 1	1.6	2.1	2.4	2.6
sample 2	1.5	2.6	2.4	2.9
sample 3	1.5	2.2	2.3	3.3
sample 4	1.9	1.7	2.8	3.7
sample 5	1.6	2.1	1.7	2.4
sample 6	1.8	2.3	2.2	2.9
Mean \pm SD	1.65 \pm 0.16 ^a	2.17 \pm 0.29 ^{ab}	2.3 \pm 0.36 ^{ab}	2.97 \pm 0.47 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S9. Fasting serum insulin (mU/L) of all animals. Value=mean \pm SD (N=6)

	VCG	PCG	MTG	NCG
sample 1	5.88	7.1	4.96	7.91
sample 2	6.07	6.66	4.97	5.94

sample 3	5.66	5.81	5.96	6.66
sample 4	6.42	5.74	6.7	6.22
sample 5	6.99	6.19	6.85	10.3
sample 6	8.33	9.34	8.42	8
Mean±SD	6.56±0.98	6.81±1.34	6.31±1.31	7.5±1.6

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S10. HOMA-IR of all animals. Value=mean ± SD (N=6)

	VCG	PCG	MTG	NCG
sample 1	1.23	4.57	1.98	7.03
sample 2	1.24	3.38	2.48	3.96
sample 3	1.23	2.69	2.38	5.45
sample 4	1.37	2.45	4.05	4.73
sample 5	1.34	2.03	3.1	9.77
sample 6	1.85	3.86	2.88	7.04
Mean±SD	1.38±0.24 ^a	3.16±0.95 ^{ab}	2.81±0.72 ^{ab}	6.33±2.09 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S11. HOMA-IS of all animals. Value=mean ± SD (N=6)

	VCG	PCG	MTG	NCG
sample 1	28.1	11.0	12.4	8.9
sample 2	29.7	13.2	9.99	8.91
sample 3	26.0	12.6	14.9	8.14
sample 4	30.1	13.5	11.1	8.19
sample 5	36.6	18.8	15.1	10.8
sample 6	37.5	22.6	24.6	9.09
Mean±SD	31.3±4.66 ^a	15.3±4.46 ^{ab}	14.7±5.27 ^{ab}	9.01±0.96 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S12. HOMA-β (%) of all animals. Value=mean ± SD (N=6)

	VCG	PCG	MTG	NCG
sample 1	97.9	12.9	18.0	9.59
sample 2	110	16.9	12.9	10.3
sample 3	80.9	16.8	21.7	8.94
sample 4	98.8	18.8	13.3	9.15
sample 5	174	31.7	20.4	11.5
sample 6	111	32.2	40.1	9.81
Mean±SD	112±32.5 ^a	21.6±8.29 ^{ab}	21.1±9.99 ^{ab}	9.88±0.92 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S13. Clinical chemistry parameters of all animals. Value=mean \pm SD (N=6)

	TG	CHO	HDL	LDL	ALT	AST	ALP
VCG	0.42 \pm 0.1 ^a	1.13 \pm 0.16 ^a	1.18 \pm 0.14	0.27 \pm 0.03 ^a	46.2 \pm 7.7 ^a	254 \pm 32.7 ^a	74.5 \pm 5.3 ^a
PCG	0.45 \pm 0.19 ^a	1.12 \pm 0.2 ^a	1.14 \pm 0.18	0.21 \pm 0.02 ^a	49.2 \pm 14.8	200 \pm 50 ^a	164 \pm 48 ^{ab}
MTG	0.4 \pm 0.26 ^a	1.1 \pm 0.44 ^a	1.08 \pm 0.16	0.22 \pm 0.07 ^a	51.7 \pm 18.7	185 \pm 40 ^a	186 \pm 60.2 ^{ab}
NCG	1.06 \pm 0.34 ^b	1.6 \pm 0.33 ^b	1 \pm 0.15	0.58 \pm 0.15 ^b	60.3 \pm 21.6 ^b	130 \pm 28.2 ^b	320 \pm 79.7 ^b

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S14. Serum NEFAs levels of all animals. Value=mean \pm SD (N=6)

	VCG	PCG	MTG	NCG
sample 1	0.57	0.33	0.42	0.74
sample 2	0.73	0.35	0.34	0.63
sample 3	0.66	0.56	0.11	0.62
sample 4	0.77	0.38	0.31	1
sample 5	0.43	0.39	0.27	1.02
sample 6	0.59	0.58	0.5	0.65
Mean \pm SD	0.63 \pm 0.12	0.43 \pm 0.11 ^{ab}	0.33 \pm 0.13 ^{ab}	0.78 \pm 0.19

a P < 0.05 compared with NCG

b P < 0.05 compared with VCG

Table S15. Taxa annotated of intestinal microflora. Value=mean \pm SD (N=6)

Taxa annotated	VCG3	PCG3	MTG3	NCG3
The total of Proteobacteria	218 \pm 91	412 \pm 118 ^{ab}	238 \pm 93.4 ^b	154 \pm 95.4
The total of Firmicutes	20099 \pm 928	20814 \pm 467	16746 \pm 1161 ^a	18422 \pm 4139
The total of Bacteroidetes	1988 \pm 988 ^b	945 \pm 302 ^a	3154 \pm 1233 ^b	696 \pm 283 ^a
The total of Actinobacteria	375 \pm 132	408 \pm 136	2584 \pm 1823 ^a	3515 \pm 4239
Verrucomicrobia Verrucomicrobiae Verrucomicrobiales Verrucomicrobiaceae Akkermansia	5 \pm 4.5	32 \pm 32	2 \pm 2.3	6 \pm 8.8
Tenericutes Mollicutes RF39	93 \pm 41.9 ^b	163 \pm 132.1 ^b	48 \pm 19.1 ^{ab}	14 \pm 7.1 ^a
ProteoGammaproteoPseudomonadales Pseudomonadaceae Pseudomonas	77 \pm 32.4 ^b	76 \pm 31.2 ^b	51 \pm 30.8	30 \pm 22.9 ^a
ProteoGammaproteoPseudomonadales Moraxellaceae Acinetobacter	21 \pm 10.8	27 \pm 13.3	15 \pm 9.2	12 \pm 7.7
ProteoGammaproteoOceanospirillales Halomonadaceae Halomonas	2 \pm 1.2	3 \pm 1.9	3 \pm 1.8	1 \pm 1
ProteoGammaproteoEnterobacteriales Enterobacteriaceae Escherichia	13 \pm 10	17 \pm 8.7	33 \pm 44.8	18 \pm 10.1
ProteoDeltaproteoDesulfovibrionales Desulfovibrionaceae	47 \pm 24.3	251 \pm 170 ^{ab}	48 \pm 15.1	69 \pm 53.2
ProteoBetaproteoBurkholderiales Comamonadaceae Tepidimonas	2 \pm 3.1	2 \pm 2	1 \pm 0.9	1 \pm 1.6
ProteoBetaproteoBurkholderiales Alcaligenaceae Sutterella	36 \pm 25.1 ^b	8 \pm 3.6 ^a	69 \pm 38.7 ^b	7 \pm 5.2 ^a
ProteoAlphaproteoSphingomonadales Sphingomonadaceae Sphingopyxis	4 \pm 1.2	11 \pm 5.9 ^{ab}	5 \pm 3.4	2 \pm 1.7
ProteoAlphaproteoSphingomonadales Sphingomonadaceae Sphingomonas	1 \pm 1.2	1 \pm 1.5	1 \pm 1.2	8 \pm 15
ProteoAlphaproteoRhizobiales Phyllobacteriaceae Aminobacter	10 \pm 5.3 ^b	9 \pm 2.3 ^b	6 \pm 3.3	5 \pm 2.9 ^a
ProteoAlphaproteoRhizobiales Brucellaceae Ochrobactrum	5 \pm 1.5	6 \pm 2.4 ^b	4 \pm 1.7	3 \pm 2.4
ProteoAlphaproteoRF32	2 \pm 4.4	1 \pm 0.8	4 \pm 4.7	1 \pm 1.6
Firmicutes Erysipelotrichi Erysipelotrichales Erysipelotrichaceae Holdemania	2 \pm 1.5	5 \pm 2.2	7 \pm 5.4	3 \pm 1.3
Firmicutes Erysipelotrichi Erysipelotrichales Erysipelotrichaceae Eubacterium	26 \pm 10.7	545 \pm 457 ^a	95 \pm 61.3	514 \pm 574
Firmicutes Erysipelotrichi Erysipelotrichales Erysipelotrichaceae Coprobacillus	52 \pm 77.3 ^b	286 \pm 262	37 \pm 16.1 ^b	635 \pm 459 ^a
Firmicutes Erysipelotrichi Erysipelotrichales Erysipelotrichaceae Clostridium	7 \pm 5.3	21 \pm 12.5 ^{ab}	24 \pm 19.5	5 \pm 4.6
Firmicutes Erysipelotrichi Erysipelotrichales Erysipelotrichaceae Allobaculum	43 \pm 11.1	133 \pm 129	97 \pm 97.1	501 \pm 735
Firmicutes Erysipelotrichi Erysipelotrichales Erysipelotrichaceae	105 \pm 43.8	183 \pm 72.1 ^b	70 \pm 34.7 ^a	66 \pm 55.2

Firmicutes Clostridia Clostridiales Veillonellaceae Phascolarctobacterium	244±137.6 ^b	56±27 ^a	374±319.8	52±34.5 ^a
Firmicutes Clostridia Clostridiales Ruminococcaceae Ruminococcus	1279±410 ^b	338±52.8 ^a	769±335	328±186 ^a
Firmicutes Clostridia Clostridiales Ruminococcaceae Oscillospira	524±169.9 ^b	342±50.1 ^a	715±650.4	262±153.5 ^a
Firmicutes Clostridia Clostridiales Ruminococcaceae Butyrivibrio	3±0.9 ^b	9±4.4 ^{ab}	2±1.5	1±1 ^a
Firmicutes Clostridia Clostridiales Ruminococcaceae	4166±626 ^b	3036±599 ^{ab}	3375±1157	2251±864 ^a
Firmicutes Clostridia Clostridiales Peptococcaceae rc4-4	8±6.7	19±9 ^a	15±10.5 ^b	7±5.6
Firmicutes Clostridia Clostridiales Mogibacteriaceae	145±27.1	308±95.8 ^{ab}	151±21.5	117±54.6
Firmicutes Clostridia Clostridiales Lachnospiraceae Ruminococcus	481±97.7 ^b	239±62.2 ^{ab}	222±46.5 ^{ab}	114±43.1 ^a
Firmicutes Clostridia Clostridiales Lachnospiraceae Roseburia	198±121.7 ^b	45±11.3 ^a	105±48.6	51±27.6 ^a
Firmicutes Clostridia Clostridiales Lachnospiraceae Dorea	1001±334 ^b	2268±636 ^a	665±167 ^{ab}	2041±540 ^a
Firmicutes Clostridia Clostridiales Lachnospiraceae Coprococcus	640±118.2 ^b	268±85.3 ^a	526±157.3	349±153.7 ^a
Firmicutes Clostridia Clostridiales Lachnospiraceae Clostridium	14±5.4	11±6.4	13±8.7	10±3.8
Firmicutes Clostridia Clostridiales Lachnospiraceae Blautia producta	366±93 ^b	1694±239 ^a	455±110 ^b	1152±471 ^a
Firmicutes Clostridia Clostridiales Lachnospiraceae Blautia	1447±270	2579±664 ^a	1263±231.1	1806±704
Firmicutes Clostridia Clostridiales Lachnospiraceae Anaerostipes	12±6.6 ^b	3±2.3 ^a	5±1.6 ^a	4±1.7 ^a
Firmicutes Clostridia Clostridiales Lachnospiraceae	2530±169	4555±989 ^a	3062±683	3983±1585
Firmicutes Clostridia Clostridiales Eubacteriaceae Anaerofustis	14±6.5	10±2.7	6±2.5	6±5.2
Firmicutes Clostridia Clostridiales Dehalobacteriaceae Dehalobacterium	8±4.6 ^b	3±2.8 ^a	3±2.2	1±0.5 ^a
Firmicutes Clostridia Clostridiales Clostridiaceae Clostridium	231±74 ^b	34±10.9 ^a	110±71.1 ^{ab}	34±20.7 ^a
Firmicutes Clostridia Clostridiales Clostridiaceae	11±4.5	17±13.5	12±7	11±3.8
Firmicutes Clostridia Clostridiales Christensenellaceae Christensenella	13±4.1	10±2.7	10±1.5	9±4.5
Firmicutes Clostridia Clostridiales Christensenellaceae	56±8.8	64±14.4	103±47.6	54±16.6
Firmicutes Clostridia Clostridiales	2682±844 ^b	1985±352 ^b	2122±536 ^b	955±456 ^a
Firmicutes Bacilli Turicibacterales Turicibacteraceae Turicibacter	25±12.6 ^b	91±46.1 ^a	95±73.2 ^a	79±51.1 ^a
Firmicutes Bacilli Lactobacillales Streptococcaceae Streptococcus	10±2.5 ^b	97±28.2 ^{ab}	20±10.7	47±34 ^a
Firmicutes Bacilli Lactobacillales Streptococcaceae Lactococcus	275±214	283±139 ^b	92±87.9	95±92.4

Firmicutes Bacilli Lactobacillales Lactobacillaceae Lactobacillus	3466±1148	1230±244 ^a	2093±1803	2867±1905
Firmicutes Bacilli Lactobacillales Enterococcaceae Enterococcus	15±5.4	31±24.8	32±31.9	11±3.7
Firmicutes Bacilli Lactobacillales Enterococcaceae	2±1.2	15±11.8 ^b	4±4.1	3±2.3
Firmicutes Bacilli Lactobacillales Carnobacteriaceae Granulicatella	1±1.4	2±2.9	1±1.2	0±0
Firmicutes Bacilli Bacillales Staphylococcaceae Staphylococcus	1±0.8	3±0.8 ^{ab}	2±0.8 ^{ab}	0±0.4
Elusimicrobia Elusimicrobia Elusimicrobiales Elusimicrobiaceae Elusimicrobium	3±0.8 ^b	1±0.8 ^a	3±4.4	0±0 ^a
Cyano4C0d-2 YS2	11±8.2 ^b	5±3.5	17±17.4	1±2.4 ^a
Bacteroidetes Bacteroidia Bacteroidales S24-7	659±241 ^b	471±258	1439±841 ^b	314±110 ^a
Bacteroidetes Bacteroidia Bacteroidales Rikenellaceae	27±18.5 ^b	7±5.8 ^a	11±7.4	3±1.5 ^a
Bacteroidetes Bacteroidia Bacteroidales Prevotellaceae Prevotella	975±664 ^b	123±27 ^a	1005±505 ^b	87±48.1 ^a
Bacteroidetes Bacteroidia Bacteroidales Porphyromonadaceae Parabacteroides	38±37.3	56±10.4	128±80.6 ^{ab}	47±31.1
Bacteroidetes Bacteroidia Bacteroidales Paraprevotellaceae Prevotella	55±18.9 ^b	31±14.3 ^b	35±25.6 ^b	6±3.7 ^a
Bacteroidetes Bacteroidia Bacteroidales Paraprevotellaceae Paraprevotella	1±2	0±0.8	3±6.9	1±0.5
Bacteroidetes Bacteroidia Bacteroidales Odoribacteraceae Odoribacter	9±6.9 ^b	1±0.8 ^a	2±3.2	0±0 ^a
Bacteroidetes Bacteroidia Bacteroidales Odoribacteraceae Butyricimonas	6±5.1	6±2.3	28±26.2	4±2.4
Bacteroidetes Bacteroidia Bacteroidales Bacteroidaceae Bacteroides	190±175	248±64.2	500±212 ^a	230±133
Bacteroidetes Bacteroidia Bacteroidales	28±27.3	4±2.7	5±3.6	5±12.2
ActinoCoriobacteriia Coriobacteriales Coriobacteriaceae Collinsella	35±14	130±70.9 ^a	29±11.6	199±212
ActinoCoriobacteriia Coriobacteriales Coriobacteriaceae Adlercreutzia	135±61.2 ^b	58±14 ^a	89±29.9	63±28 ^a
ActinoCoriobacteriia Coriobacteriales Coriobacteriaceae	150±59.8	68±16.2 ^a	327±96.3 ^a	311±305
ActinoActinoBifidobacteriales Bifidobacteriaceae Bifidobacterium	37±30.4 ^b	117±63.6 ^{ab}	2124±1726 ^a	2924±4347 ^a
ActinoActinoBifidobacteriales Bifidobacteriaceae	0±0.4 ^b	3±1.2 ^a	0±0.5 ^b	2±1.4 ^a
ActinoActinoActinomycetales Nocardioidaceae Aeromicrobium	4±1.6 ^b	3±1.9	2±1.4	2±1.5 ^a
ActinoActinoActinomycetales Nocardiaceae Rhodococcus	2±1.8	1±0.5	2±0.5	1±0.8
ActinoActinoActinomycetales Micrococcaceae Rothia	12±3.7	24±18.7	9±4.9 ^a	11±5.9
ActinoActinoActinomycetales Brevibacteriaceae Brevibacterium	2±1.5	4±2.3	2±1.3	2±2

a, $P < 0.05$ compared with VCG

b, $P < 0.05$ compared with NCG