

## Supplementary figures

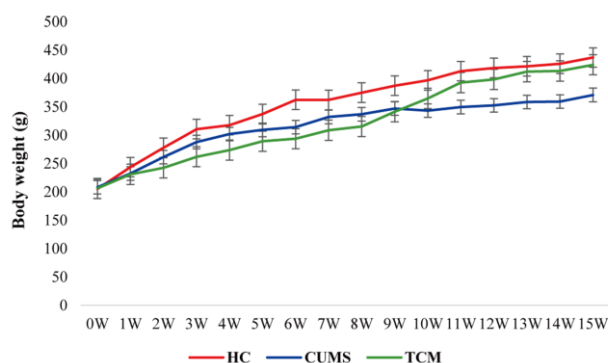


Figure S1. The change tendency of body weight among three groups. All data are expressed as mean  $\pm$  standard error of the mean (SEM) (HC: n=12; CUMS: n=6; TCM: n=19).

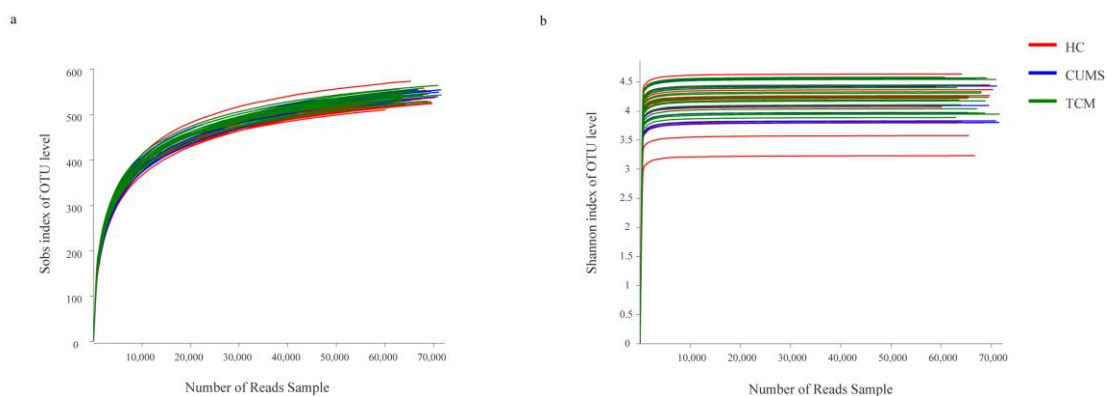


Figure S2. Bacterial rarefaction curves based on (a) Shannon index, and (b) Sobs index. Each sample was distinguished by different colors of lines.

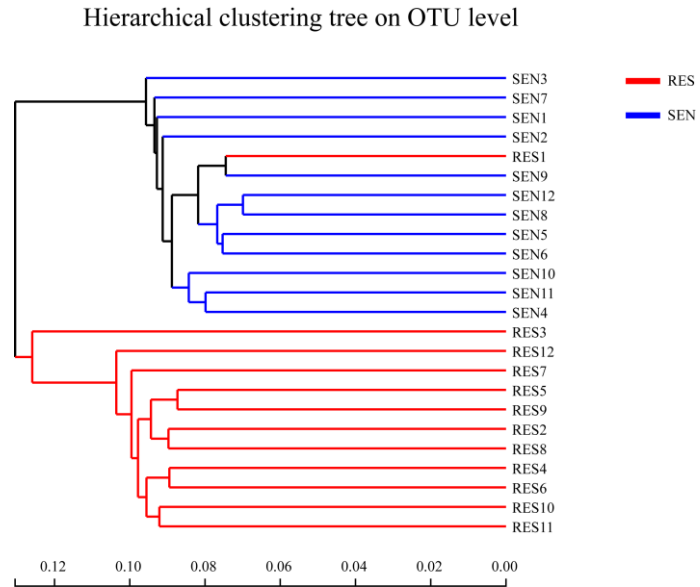


Figure S3. Hierarchical clustering tree analysis was performed at operational taxonomic unit (OTU) level based on bray\_curtis algorithm for SEN and RES group. (SEN: rats are sensitive to CUMS; RES: rats are resistant to CUMS)

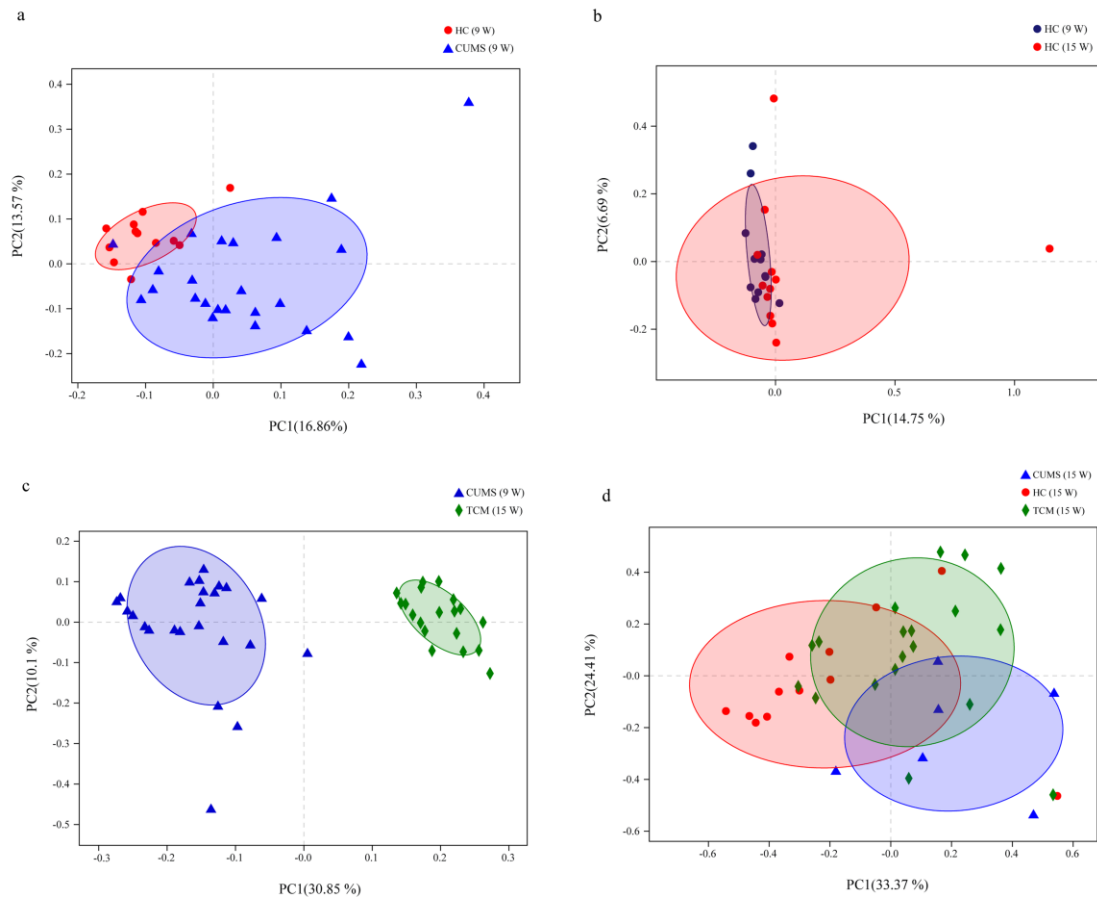


Figure S4. Principal coordinate analysis (PCoA) was performed at operational taxonomic unit (OTU) level based on bray\_curtis algorithm for all samples. Each group is represented in a different color and shape. **(a)** Scatterplot from PCoA showed a clear separation of HC group from CUMS group in Week 9 ( $p < 0.05$ , analysis of similarity (ANOSIM)); Principal coordinates 1 and 2 (PC1 and PC2) represent 16.86%, 13.57% of the variance, respectively (x and y axis) (HC (9 W):  $n=12$ ; CUMS (9 W):  $n=24$ , representative). **(b)** Scatterplot from PCoA presented a similar bacterial composition between HC (9 W) and HC (15 W) (HC (9 W):  $n=12$ ; HC (15 W):  $n=12$ ). **(c)** PCoA Scatterplot identified bacteria compositional difference between pre- and post-TCM treatment group ( $p < 0.05$ , analysis of similarity (ANOSIM)); Principal coordinates 1 and 2 (PC1 and PC2) represent 30.85%, 10.1% of the variance, respectively (x and y axis) (CUMS (9 W):  $n=24$ ; TCM (15 W):  $n=19$ ). **(d)** Scatterplot from PCoA identified bacteria compositional difference among three groups ( $p < 0.05$ , analysis of similarity (ANOSIM)) in Week 15; Principal coordinates 1 and 2 (PC1 and PC2) represent 33.7%, 24.41% of the variance, respectively (x and y axis) (HC:

n=12; CUMS: n=6; TCM: n=19)

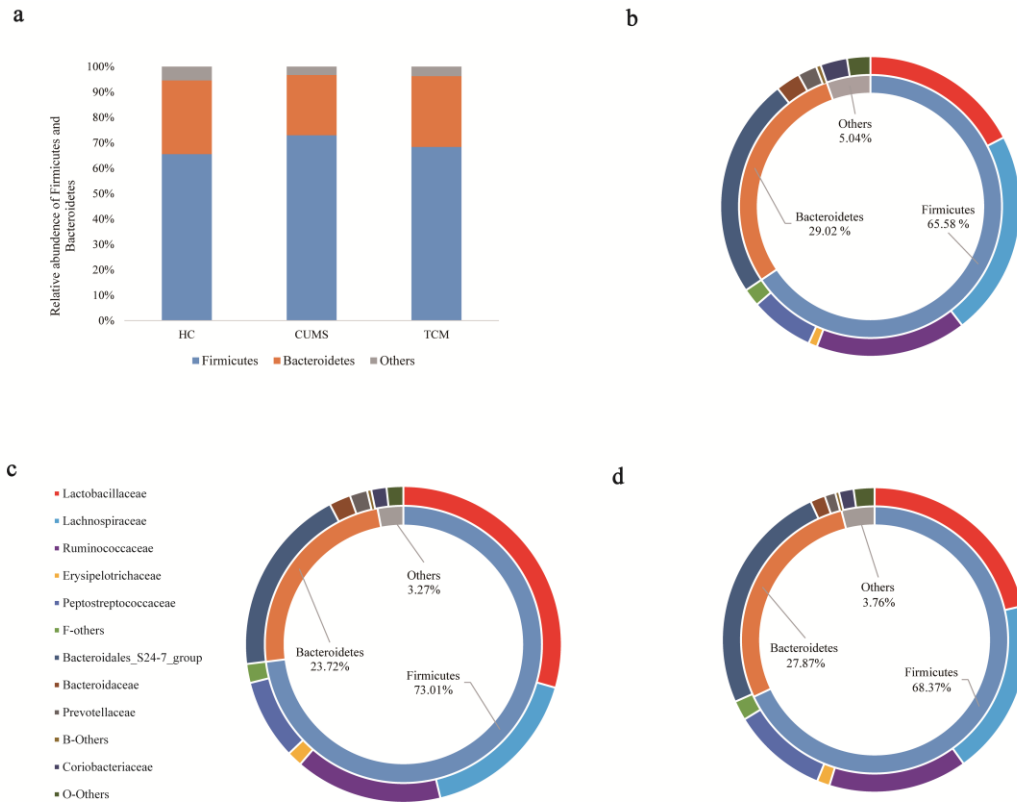


Figure S5. The main phylum and families of representative samples detected in three groups (HC: n=6; CUMS: n=5; TCM: n=6, representative). **(a)** The main phylum detected in the fecal samples. **(b)** The main families detected in the HC group. **(c)** The main families detected in the CUMS group. **(d)** The main families detected in the TCM group.

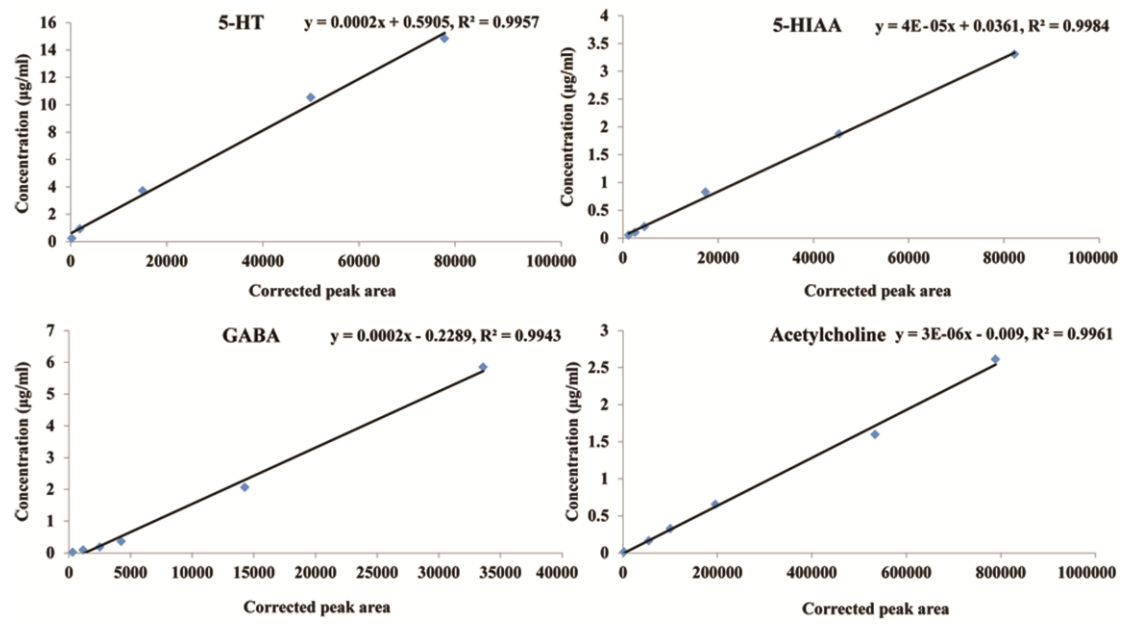


Figure S6. Calibration curves of each external standard

## Supplementary tables

**Table S1. The composition of TCM herbal formula.**

<b>Herbs</b>	<b>Amount (g)</b>
<i>Bupleurum chinense</i> DC.	30
<i>Angelica sinensis</i> (Olive.) Diel	30
<i>Paeonia lactiflora</i> Pall	30
<i>Atractylodes macrocephala</i> Koidz.	30
<i>Poria cocos</i> (Schw.)Wolf	30
<i>Glycyrrhiza uralensis</i> Fisch.	15
<i>Zingiber officinale</i> Rosc.	10
<i>Mentha haplocalyx</i> Briq.	10

**Table S2. Contents of active ingredient in Component I and Component II for this formula.**

<b>Component</b>	<b>Active ingredients</b>	<b>Methods</b>	<b>Amounts (mg/g)</b>
<b>Component I</b> (164.6 mg/kg/d)	Ferulic acid	HPLC	0.05
	Paeoniflorin	HPLC	4.88
	Aurantiamarin	HPLC	5.14
<b>Component II</b> (31.25 mg/kg/d)	Polyphenols	UV	500
	Total Catechins	HPLC	440
	Epigallocatechin gallate (EGCG)	HPLC	180
	Caffeine	HPLC	80

**Table S3. Species richness and diversity of three groups.**

Group	Numbers of species observed	Community richness		Community diversity	
		Chao 1	Ace	Simpson	shannon
HC	501.00 ± 3.8471	511.48 ± 11.2696	509.20 ± 6.1170	0.9573 ± 0.0235	6.3081 ± 0.3526
CUMS	506.80 ± 12.1326	523.09 ± 14.0112	523.64 ± 15.9834	0.9312 ± 0.0266	5.8450 ± 0.4439
TCM	506.67 ± 11.1833	519.96 ± 10.0392	516.19 ± 12.4156	0.9493 ± 0.0202	6.0899 ± 0.3358

**Note:** Mean values ± standard deviation are listed (HC: n=6; CUMS: n=5; TCM: n=6, representative).

**Table S4. Relative abundance of significant different key taxa among three groups.**

Taxa	HC	CUMS	TCM	<i>p</i> -Value			
				HC vs CUMS	CUMS vs TCM	HC vs TCM	
Phylum	<i>Firmicutes</i>	65.58 ± 1.6227	73.01 ± 2.8019	68.37 ± 1.7652	0.048	/	0.013
	<i>Bacteroidetes</i>	29.02 ± 2.0397	23.72 ± 2.7969	27.87 ± 1.8503	0.004	/	0.037
	<i>Actinobacteria</i>	3.33 ± 0.7526	1.76 ± 0.3288	2.01 ± 0.1938	0.049	/	/
	<i>Deferribacteres</i>	0.1236 ± 0.0533	0.0146 ± 0.0027	0.0314 ± 0.0036	0.037	/	/
Class	<i>Bacilli</i>	18.22 ± 3.6720	29.99 ± 4.3518	22.08 ± 3.2883	0.046	/	/
	<i>unidentified_Deferribacteres</i>	0.1236 ± 0.0533	0.0146 ± 0.0027	0.0314 ± 0.0036	0.037	/	/
Order	<i>Lactobacillales</i>	17.77 ± 3.6628	29.78 ± 4.3522	21.78 ± 3.2771	0.042	/	/
	<i>Bacillales</i>	0.443 ± 0.0699	0.2129 ± 0.0107	0.3084 ± 0.0314	0.005	/	/
	<i>Lactobacillaceae</i>	17.39 ± 3.6300	29.42 ± 4.3322	21.41 ± 3.2594	0.041	/	/
Family	<i>Clostridiaceae_1</i>	0.3330 ± 0.0574	0.7393 ± 0.0852	0.7486 ± 0.1009	0.004	/	0.003
	<i>Staphylococcaceae</i>	0.4315 ± 0.0715	0.1984 ± 0.0095	0.2915 ± 0.0316	0.005	/	/
	<i>Family_XIII</i>	0.4280 ± 0.0449	0.3083 ± 0.0225	0.3718 ± 0.0364	0.043	/	/
Family	<i>Deferribacteraceae</i>	0.1236 ± 0.0533	0.0146 ± 0.0027	0.0314 ± 0.0036	0.037	/	/
	<i>Streptococcaceae</i>	0.191 ± 0.0253	0.1233 ± 0.0112	0.1436 ± 0.0122	0.021	/	/
Genus	<i>Lactobacillus</i>	17.39 ± 3.6300	29.42 ± 4.3322	21.41 ± 3.2594	0.041	/	/
	<i>Enterorhabdus</i>	1.40 ± 0.2608	0.7947 ± 0.1655	0.6805 ± 0.0664	0.039	/	0.013
	<i>Ruminococcus_2</i>	0.2749 ± 0.0911	0.7687 ± 0.1992	0.3744 ± 0.0678	0.014	0.042	/
	<i>Roseburia</i>	0.9878 ± 0.0658	0.7114 ± 0.0242	0.9400 ± 0.0765	0.009	0.025	/
	<i>Clostridium sensu stricto_1</i>	0.3330 ± 0.0574	0.7393 ± 0.0852	0.7486 ± 0.1009	0.004	/	0.003
	<i>Lachnospiraceae_UCG-001</i>	0.4970 ± 0.0815	0.2386 ± 0.0405	0.2575 ± 0.0413	0.010	/	0.011

**Note:** Key taxa were identified applying the metagenomic biomarker discovery approach of one-way ANOVA and only the *p*-value less than 0.05 are shown. Data are expressed as mean ± Standard error of the mean (SEM). Abbreviations: HC (health control); CUMS (chronic unpredictable mild stress); TCM (traditional Chinese medicine).