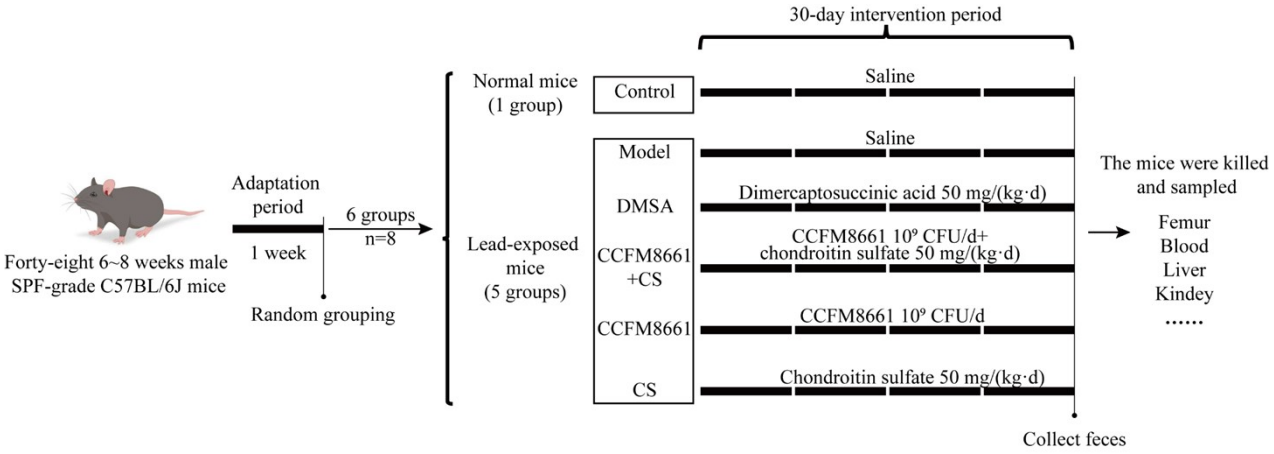
Supplementary Materials

Animal experimental design

Forty-eight 6~8 weeks male SPF-grade C57BL/6J mice were housed in an experimental environment of 24±1 °C, 50±10% humidity, and 12 h alternating light and dark. All animals were randomly grouped into six groups (Control, model, DMSA, CCFM8661+CS, CCFM8661, and CS groups) after a one-week acclimatization phase and treated with Pb exposure, probiotic intervention, etc. A detailed illustration is shown in Figure S1. The control group was not exposed to lead; the other five groups used aqueous lead acetate containing 1 g/L of lead ions as their daily drinking water to construct the lead exposure model. The control and model groups were gavaged with saline, and the other four groups were gavaged with the corresponding agents.



1. All interventions were administered by gavage, and the volume of gavage was 0.2mL.
2. Mice were given a lead acetate solution with a concentration of 1.0 g/L of lead ions per day to construct a lead-exposed mouse model.

Figure S1 Schematic diagram of animal experiment design

Microbial gene sequencing

Total bacterial DNA in the feces was extracted from the samples following the manufacturer's protocol (Fast DNA Stool Kit, MP Biomedicals, CA, USA), and the extracted DNA was amplified in the V3-V4 region of the 16S rRNA (universal primers, 341F/806R). The amplification comprised an initial denaturation stage at 95 °C for 5 minutes, followed by 30 cycles of 95 °C for 30 seconds, 52 °C for 30 seconds, and 72 °C for 30 seconds, and a final extension step at 72 °C. After cycling, a final extension at 72 °C for 5 minutes was performed, and the

reaction was then kept at 4 °C. The obtained DNA was recovered and purified using the TIANgel Mini Purification Kit (TIANGEN, Beijing, China). DNA was quantified and pooled in equal concentrations following the instructions for the Qubit dsDNA Assay Kit (Life Technologies, Carlsbad, CA, USA). Libraries were generated using the TruSeq DNA LT Sample Preparation Kit (Illumina, San Diego, CA, USA), and samples were barcoded and paired-end sequenced on the Illumina MiSeq PE300 platform following the manufacturer's protocol.

Table S1 Differences in fecal metabolites between the Control and Model groups.

Number	Metabolite	VIP	FC	*P*	Change
1	Cinnamoylglycine	1.36	43.33	*	↑
2	Hippuric acid	1.32	31.88	**	↑
3	Equol	2.05	26.92	****	↑
4	4-Acetamidobenzoic acid	1.32	8.36	*	↑
5	3-Methoxysalicylic acid	1.57	4.03	**	↑
6	6-Methylnicotinamide	1.52	3.93	***	↑
7	Adenine	1.53	3.79	**	↑
8	Protocatechuic acid	1.5	3.78	**	↑
9	(+/-)12(13)-DiHOME	1.86	3.03	****	↑
10	L-(+)-Arginine	1.29	2.68	**	↑
11	(+/-)9,10-dihydroxy-12Z-octadecenoic acid	1.83	2.66	****	↑
12	Genistein	1.51	2.54	***	↑
13	Glycitein	1.57	2.52	***	↑
14	DL-Carnitine	1.69	2.51	**	↑
15	Linoleoyl Ethanolamide	1.75	2.50	**	↑
16	13(S)-HOTrE	1.69	2.47	****	↑
17	DL-Lactic Acid	1.35	2.43	***	↑
18	9S,13R-12-Oxophytodienoic acid	1.27	2.38	**	↑
19	Gluconic acid	1.54	2.33	**	↑
20	Succinic acid	1.21	2.26	**	↑
21	2-Hydroxyhippuric acid	1.37	2.20	*	↑
22	9-Oxo-10(E),12(E)-octadecadienoic acid	1.52	2.14	***	↑
23	Ferulic acid	1.47	2.12	***	↑
24	Vanillin	1.35	2.12	**	↑
25	Oleamide	1.52	2.1	**	↑
26	1,6-Hydroxyhexadecanoic acid	1.72	2.09	****	↑
27	DL-Stachydrine	1.29	2.09	*	↑
28	2,4-Dihydroxybenzoic acid	1.1	2.07	*	↑
29	Choline	1.45	2.06	***	↑
30	Taurochenodeoxycholic acid	1.34	0.48	**	↓
31	Heptanoic acid	1.27	0.47	*	↓
32	2,4-Quinolinediol	1.13	0.46	*	↓
33	Kynurenic acid	1.31	0.45	*	↓

Number	Metabolite	VIP	FC	*P*	Change
34	N8-Acetylspermidine	1.29	0.45	*	↓
35	Uridine	1.44	0.45	**	↓
36	4-Indolecarbaldehyde	1.52	0.44	****	↓
37	Valeric acid	1.37	0.44	*	↓
38	3-(4-Hydroxyphenyl)propionic acid	1.6	0.41	**	↓
39	Hypoxanthine	1.62	0.41	**	↓
40	Pentadecanoic acid	1.42	0.41	**	↓
41	4-Pyridoxic acid	1.04	0.38	*	↓
42	Docosahexaenoic acid ethyl ester	1.42	0.38	**	↓
43	Phenylacetaldehyde	1.68	0.38	***	↓
44	3-Coumaric acid	1.54	0.37	**	↓
45	11(Z),14(Z)-Eicosadienoic acid	1.84	0.36	***	↓
46	DL-4-Hydroxyphenyllactic acid	1.45	0.36	***	↓
47	Guanine	1.71	0.36	***	↓
48	Hexanoic acid	1.47	0.35	**	↓
49	Thymine	1.55	0.34	**	↓
50	Isobutyric acid	1.21	0.33	*	↓
51	2-Deoxyinosine	1.55	0.31	**	↓
52	4-Methylphenol	1.6	0.29	****	↓
53	1-[4-hydroxy-5-(hydroxymethyl)tetrahydrofuran-2-yl]pyrimidine-2,4(1H,3H)-dione	1.55	0.28	**	↓
54	2-Anisic acid	1.76	0.26	****	↓
55	4-Coumaric acid	1.42	0.23	**	↓
56	Dodecanedioic acid	1.42	0.1	**	↓

Note: VIP: Variable Importance in the Projection. FC: Flod Change, in the comparison of Control group and Model group, the data of Model group is taken as the baseline; ↑ represents the increase of substance content, ↓ represents the decrease of substance content. *, **, ***, and **** correspond to $P < 0.05$, < 0.01, < 0.001, and < 0.0001, respectively.

Table S2 Differences in fecal metabolites between the Model and CCFM8661-CS groups.

Number	Metabolite	VIP	FC	*P*	Change
1	Hydrocinnamic acid	1.98	41.30	****	↑
2	Cinnamoylglycine	1.43	31.24	**	↑
3	Equol	2.35	28.77	****	↑
4	Hippuric acid	1.39	20.13	**	↑
5	5-Hydroxyindole-3-acetic acid	2.07	12.64	****	↑
6	4-Hydroxybenzaldehyde	1.70	7.91	****	↑
7	4-Acetamidobenzoic acid	1.41	7.61	**	↑
8	Cyclopentylacetic acid	1.89	6.33	****	↑
9	Pantothenic acid	1.89	5.54	**	↑
10	4-Indolecarbaldehyde	1.93	4.57	****	↑
11	2,4-Quinolinediol	1.83	3.10	****	↑
12	L-(+)-Arginine	1.28	2.93	*	↑
13	S-Adenosylmethionine	1.34	2.86	*	↑

Number	Metabolite	VIP	FC	*P*	Change
14	(8aR,12S,12aR)-12-Hydroxy-4-methyl-4,5,6,7,8,8a,12,12a-octahydro-2H-3-benzoxecine-2,9(1H)-dione	1.52	2.43	**	↑
15	6-Methylnicotinamide	1.25	2.26	*	↑
16	Ethoxyquin	2.11	2.26	***	↑
17	Succinic acid	1.29	2.21	*	↑
18	Indole-3-lactic acid	1.67	0.39	*	↓
19	2-Hydroxycaproic acid	1.37	0.37	**	↓
20	4-Hydroxybenzoic acid	1.79	0.35	**	↓
21	Daidzein	1.95	0.32	***	↓
22	Guanine	1.43	0.30	**	↓
23	Hypoxanthine	1.56	0.30	**	↓
24	N-Acetyl-L-methionine	1.79	0.30	**	↓
25	Phenylacetaldehyde	2.43	0.30	****	↓
26	5-Hydroxylysine	2.33	0.28	****	↓
27	Guanosine	1.36	0.21	**	↓
28	DL-4-Hydroxyphenyllactic acid	1.85	0.19	*	↓
29	Phenol	1.92	0.19	***	↓
30	Dodecanedioic acid	1.68	0.16	*	↓
31	4-Toluic acid	2.17	0.14	****	↓
32	3-(4-Hydroxyphenyl)propionic acid	2.48	0.10	****	↓
33	4-Coumaric acid	1.94	0.09	****	↓

Note: VIP: Variable Importance in the Projection. FC: Flod Change, in the comparison of CCFM8661+CS group and Model group, the data of Model group is taken as the baseline; ↑ represents the increase of substance content, ↓ represents the decrease of substance content. *, **, ***, and **** correspond to $P < 0.05$, < 0.01, < 0.001, and < 0.0001, respectively.

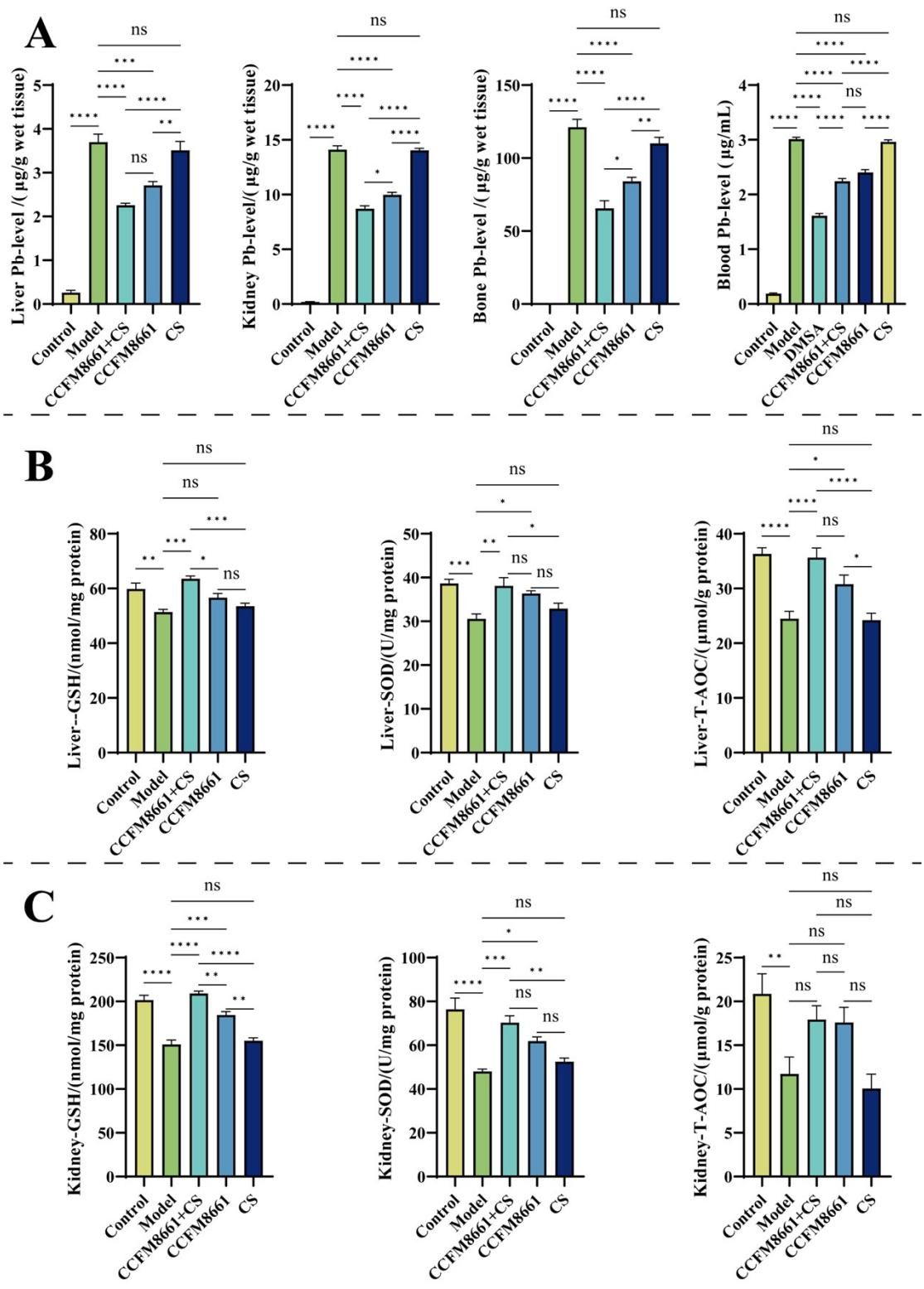


Figure S2 Effectiveness of CCFM8661+CS versus CCFM8661 or chondroitin sulphate alone. (A) Lead content in liver, kidney, bone and blood. (B) Liver oxidation index. (C) Renal oxidation index.

The liver, kidney and bone tissues of lead-exposed mice were enriched with significant amounts of lead, and their lead contents reached 3.70, 14.11 and 121.20 mg/g wet tissue,

respectively. After CCFM8661 + CS intervention, a significant decrease of lead was observed in the liver, kidney and bone tissues of the mice, and the tissue lead contents were 2.26, 8.72 and 65.57 mg/g wet tissue, respectively. In CCFM8661 alone, the liver, kidney and bone tissue lead levels were 2.71, 9.97 and 84.09 mg/g wet tissue, respectively; and in CS alone, the liver, kidney and bone tissue lead levels were 3.51, 14.04 and 110.10 mg/g wet tissue, respectively.

In terms of the reduction values of tissue lead content, the reduction values of CCFM8661 + CS were greater than the sum of the reduction values of CCFM8661 and CS respectively, i.e., the lead content was (Model group - CCFM8661 + CS group) > ((Model group - CCFM8661 group) + ((Model group - CS group)). These results indicate that the intervention of CCFM8661 + CS can significantly reduce lead accumulation in the tissues of lead-exposed mice, and the synergistic effect of the two when used together is better than that of CCFM8661 or CS alone, or the effect of the two alone in combination.

Similar results were presented in the oxidative indices of liver and kidney, which also indicated that the combination of CCFM8661+CS was more effective than CCFM8661 alone or chondroitin sulphate alone.

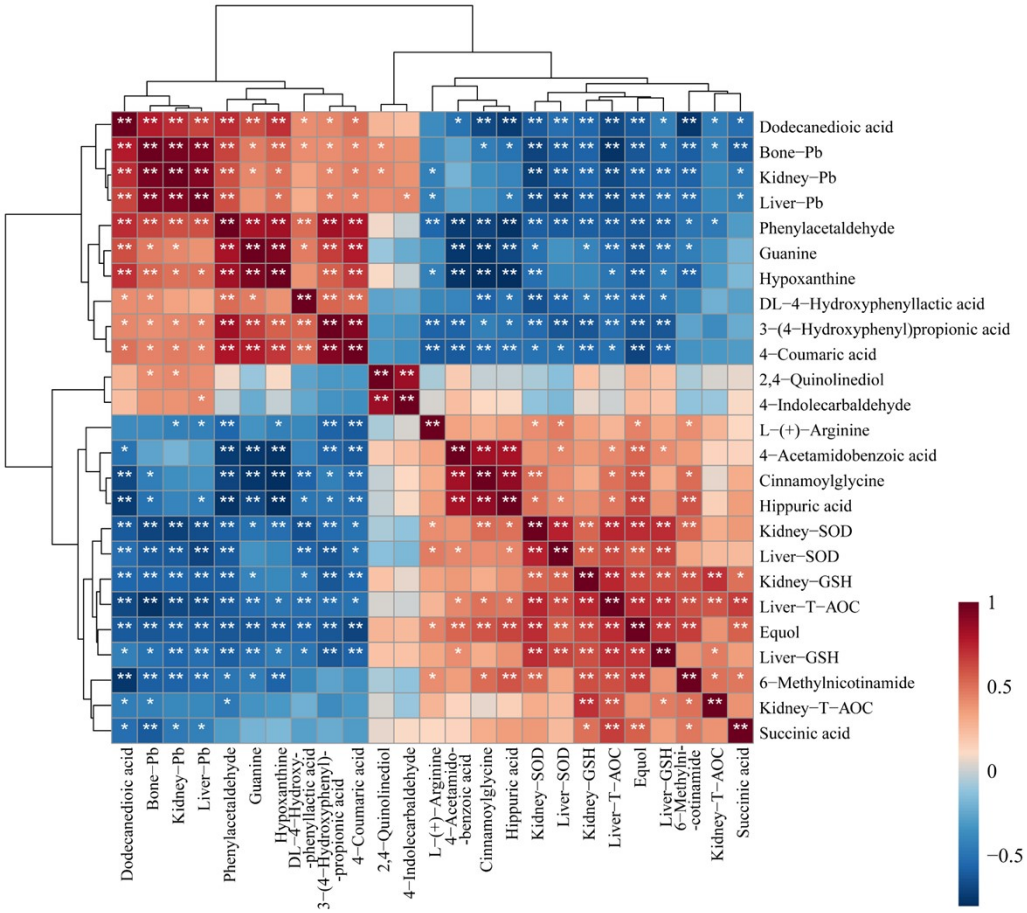


Figure S3 Spearman correlation analysis of key differential metabolites with tissue lead levels and antioxidant indices. The correlation analysis are analyzed in the online website: https://www.omicstudio.cn/tool.

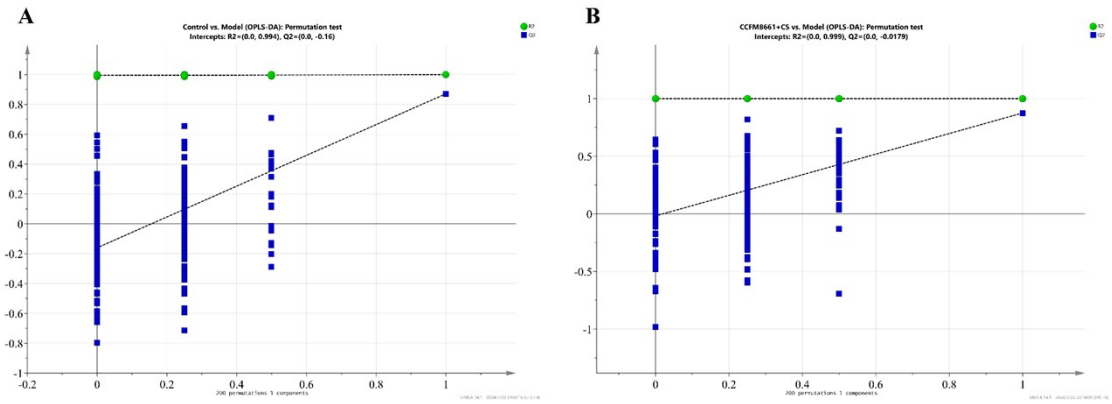


Figure S4 Permutation test results of OPLS-DA. (A) Results of the control versus model group, Intercepts: R^2=(0,0.994), Q^2=(0,-0.16). (B) Results of the CCFM8661+CS versus model group, Intercepts: R^2=(0,0.999), Q^2=(0,-0.0179).

The results of the perutation test demonstrate that the R2 tends towards 1. In addition, the blue Q2 values on the left are lower than the original points on the right. Also, the blue regression line for the Q2-points intersects the vertical axis (left side) at or below zero. The above results show the reliability of the OPLS-DA model.