

Electronic Supplementary Information:

Stabilization of hypoxia-inducible factor 1 α and regulation of specific gut microbes

by EGCG contribute to the alleviation of ileal barrier disorder and obesity

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Table S1 The formulas of the low-fat control diet and high-fat diet

Product #	D12450J		D12492	
	gm%	kcal%	gm%	kcal%
Protein	19.2	20	26	20
Carbohydrate	67.3	70	26	20
Fat	4.3	10	35	60
Total		100		100
kcal/gm	3.85		5.24	
Ingredient	gm	kcal	gm	kcal
Casein,80 Mesh	200	800	200	800
L-Cystine	3	12	3	12
Corn Starch	506.2	2024.8	0	0
Maltodextrin 10	125	500	125	500
Sucrose	68.8	275.2	68.8	275.2
Cellulose, BW200	50	0	50	0
Soybean Oil	25	225	25	225
Lard	20	180	245	2205
Mineral Mix S10026	10	0	10	0
DiCalcium Phosphate	13	0	13	0
Calcium Carbonate	5.5	0	5.5	0
Potassium Citrate,1H2O	16.5	0	16.5	0
Vitamin Mix V10001	10	40	10	40
Choline Bitartrate	2	0	2	0
FD&C Yellow Dye #5	0.04	0	0	0
FD&C Blue Dye #1	0.01	0	0.05	0
Total	1055.05	4057	773.85	4057

Table S2 Primer sequences designed for mice

Gene	Forward primer (5'→3')	Reverse primer (5'→3')
HSL	GCTAGCCAGGCTCATCTCCT	GTTCTTGAGGTAGGGCTCGT
ACOX	CTATGGGATCAGCCAGAAAGG	AGTCAAAGGCATCCACCAAAG
Leptin	CCTGTGGCTTTGGTCCATCTG	AGGCAAGCTGGTGAGGATCTG
ACC	GGCAGCAGTTACACCACATAC	TCATTACCTCAATCTCAGCATAGC
PGC1 α	AGCCGTGACCACTGACAACGAG	GCTGCATGGTTCTGAGTGCTAAG
TNF α	AATGGCCTCCCTCTCATCAG	CCACTTGGTGGTTTGCTACG
IL-6	ACTTCCATCCAGTTGCCTTCTTG	TGTTGGGAGTGGTATCCTCTGTG
IL-1 β	AAGGGCTG TTCCAAACCTTTGAC	TGCCTGAAGCT TTGTTGATGTGC
Reg3 γ	CCATCTTCACGTAGCAGC	CAAGATGTCCTGAGGGC
Angio4	TGGCCAGCTTTGGAATCACTG	GCTTGGCATCATAGTGCTGACG
α -defensin	GGTGATCATCAGACCCAGCATCAGT	AAGAGACTAAAAGTGGAGGAGCAGC
ZO-1	GGGGCCTACACTGATCAAGA	TGGAGATGAGGCTTCTGCTT
Occludin	ACGGACCCTGACCACTATGA	TCAGCAGCAGCCATGTACTC
JAM-A	GCCAGATCACAGCTCCCTAT	ACTGATCGTCGGCTTGGATG
Mucin2	ACCTGGAAGGCCCAATCAAG	CTCAGCGTAGTTGGCACTCT
HIF1 α	TTAAAGCCAACTCTTTGCTCCG	ATGGGGGCATTACCAGACAG
β -actin	ACAGCAGTTGGTTGGAGCAA	ACGCGACCATCCTCCTCTTA

Primer sequences designed for Caco-2

Gene	Forward primer (5'→3')	Reverse primer (5'→3')
ZO-1	GGGTAACGCCATCCTCTGAA	CTGGTCCTCCTTTCAGCACA
Occludin	TCAGGGAATATCCACCTATCACTTCAG	CATCAGCAGCAGCCATGTACTCTTCAC
Claudin	GCGCGATATTTCTTCTTGCAGG	TTCGTACCTGGCATTGACTGG
JAM-A	ATAGCCGAGGCCACTTTGAC	TTCTCCTTCACTTCGGGCAC
GAPDH	CAACGGATTTGGTCGTATTGGG	AAGGGGTCATTGATGGCAAC

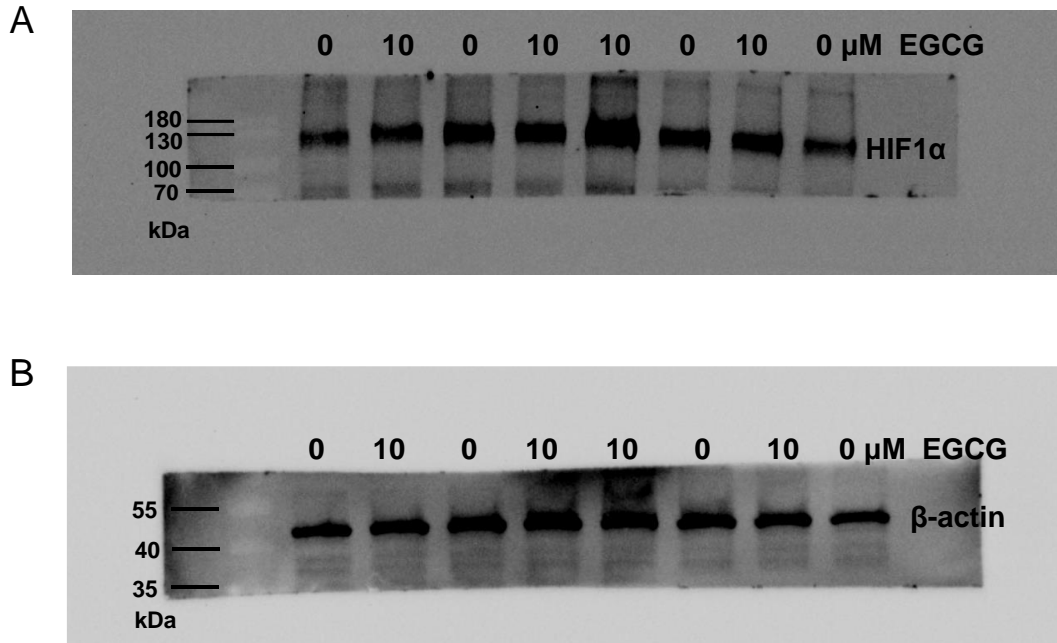


Fig. S1 Immunoblot analysis of HIF1α protein expression in cell monolayers. The Western blot was loaded with samples incubated with 0 or 10 μM EGCG (four replicates each) in the following order: 0, 10, 0, 10, 10, 0, 10, 0.

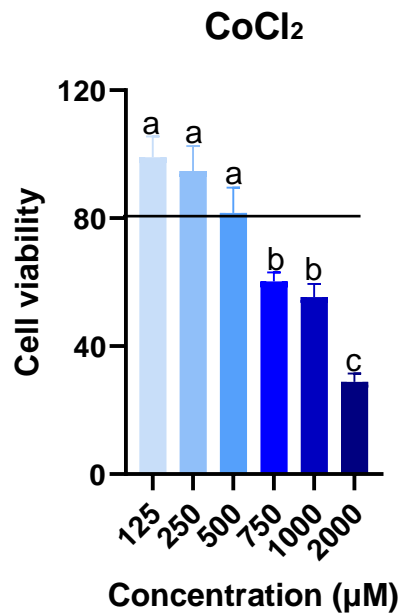


Fig. S2 Assessment of the different concentrations of CoCl₂ on Caco-2 cell viability by the CCK-8 assay

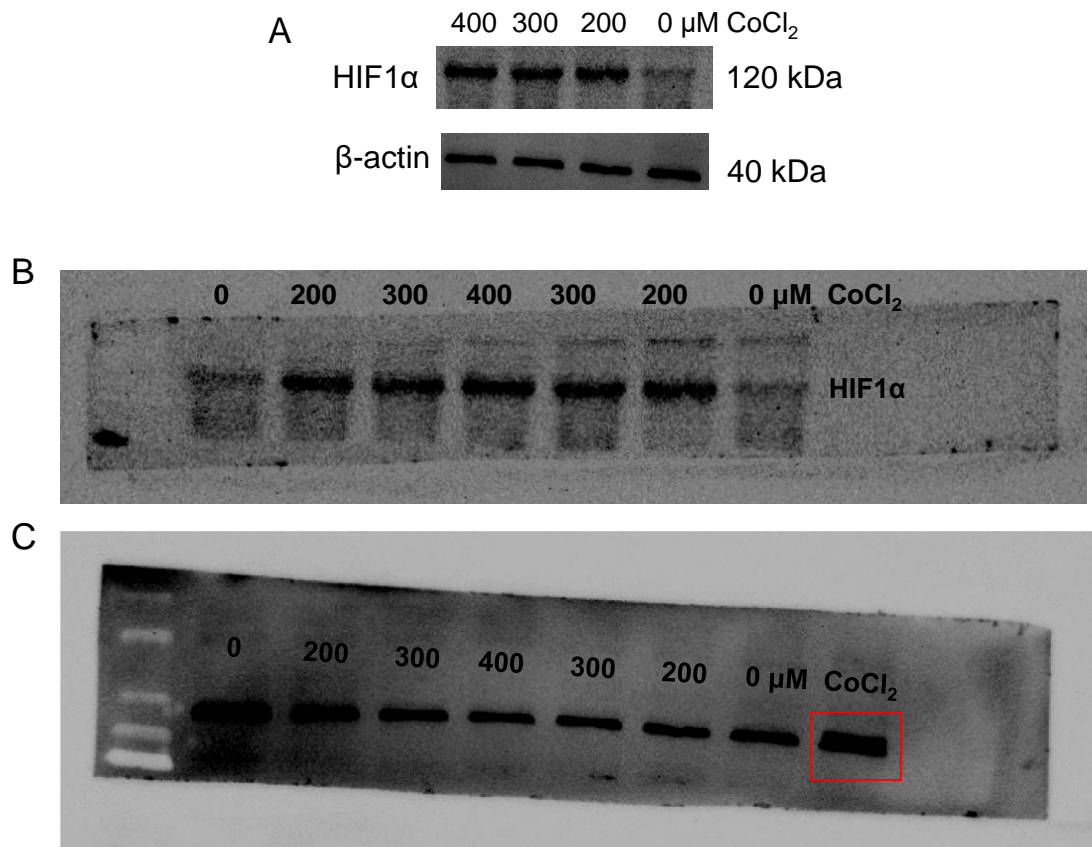


Fig. S3 Stabilizing effect of different concentrations of CoCl_2 treatment on HIF-1 α protein expression. The Western blot was loaded with samples incubated with 0, 200, 300 and 400 μM EGCG (two replicates for 0, 200 and 300 μM) in the following order: 0, 200, 300, 400, 300, 200, 0 μM . The band in the red box is not relevant to this experiment.