**Supplementary Materials**

**for**

# An Association between Decreased Small Intestinal RNA Modification and Disturbed Glucagon-like Peptide-1 Secretion under High-Fat Diet Stress

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**Content:**

**Table S1 Ingredients of normal-chew and high-fat diet**

| Serial number | Normal-chew Diet (D12450B) | | High-fat Diet (D12492) | |
|---|---|---|---|---|
| Macronutrient | **g%** | **Kcal%** | **g%** | **Kcal%** |
| Protein | 19.2 | 20.0 | 26.2 | 20.0 |
| Carbohydrate | 67.3 | 70.0 | 26.3 | 20.1 |
| Fat | 4.3 | 10.0 | 34.9 | 59.9 |
| Total | - | 100.0 | - | 100.0 |
| Kcal/g* | 3.85 | | 5.24 | |
| | | | | |
| Ingredient | **g** | **Kcal** | **g** | **Kcal** |
| Casein, 80 mesh meshes | 200 | 800 | 200 | 800 |
| DL-Methionine | 3 | 12 | 3 | 12 |
| Corn starch | 315 | 1260 | 0 | 0 |
| Maltodextrin 10 | 35 | 140 | 125 | 500 |
| Sucrose | 350 | 1400 | 68.8 | 275.2 |
| Cellulose BW20 | 50 | 0 | 50 | 0 |
| Soybean oil | 25 | 225 | 25 | 225 |
| Lard | 20 | 180 | 245 | 2205 |
| Complex mineral | 10 | 0 | 10 | 0 |
| Dicalcium phosphate | 13 | 0 | 13 | 0 |
| Calcium carbonate | 5.5 | 0 | 5.5 | 0 |
| Potassium citrate, monohydrate | 16.5 | 0 | 16.5 | 0 |
| Multivitamin V10001 | 10 | 40 | 10 | 40 |
| Choline Bitartrate | 2 | 0 | 2 | 0 |
| FD&C Yellow Dye #5 Pigment | 0.05 | 0 | 0 | 0 |
| FD&C Red Dye #40 Pigment | 0 | 0 | 0 | 0 |
| FD&C Blue Dye #1 | 0 | 0 | 0.05 | 0 |
| Total | **1055.05** | **4057** | **773.85** | **4057** |

* Nutrient to calorie ratio (Kcal/g) was used to measure the amount of a specific nutrient (or group of nutrients) in a diet per calorie.

**Table S2 Sequence of the used primers**

| Gene name | Primers |
|---|---|
| *gcg* | Forward: AGCGACTACAGCAAATAC<br>Reverse: GTTCCTCTTGGTGTTCAT |
| *pc3* | Forward: AAGTGGTCGTTACAGATGTGA<br>Reverse: AGAATCCTTGGTCCGTGTT |
| *mettl3* | Forward: ATCCAGGCCCATAAGAAACAG<br>Reverse: CTATCACTACGGAAGGTTGGG |
| *mettl14* | Forward: CTGAGAGTGCGGATAGCATTG<br>Reverse: GAGCAGATGTATCATAGGAAGCC |
| *wtap* | Forward: GCCCCAACGTTTAAGTGCAG<br>Reverse: AGCATTCGACACTTCGCCAT |
| *gapdh* | Forward: TTCCAGTATGACTCTACCCACGGCA<br>Reverse: GCACCAGCATCACCCCATTTG |

**Figure S1 Typical mass chromatogram of Adenosine, m6A, Cytidine and m5C.**

(A) Total ion chromatography (TIC) of the digested samples; (B-E) Selected ion recording (SIR) of A, m6A, C, m5C.

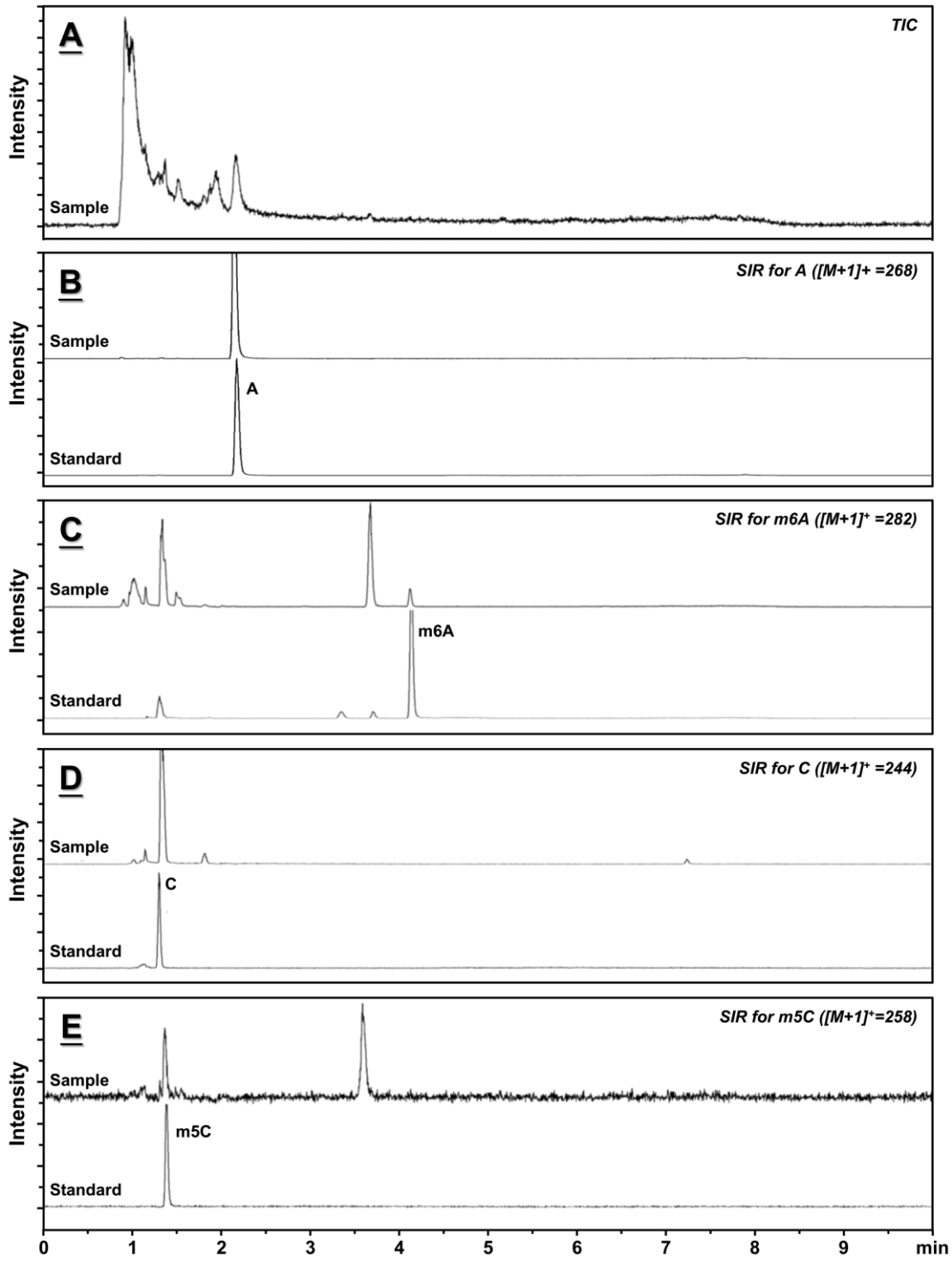

**Figure S2 High-fat diet induced a typical obesity on mice (n =6).**

(A) body weight; (B) fat mass; (C) adipocyte size of epididymis fat; (D) liver mass; (E) Lee's index; (F) fasting blood glucose; (G) fasting serum insulin; (H) homeostasis model assessment (HOMA)-insulin resistance (IR) index; (I) HOMA-insulin sensitivity (IS) index; Blood contents of (J) total cholesterol (TC), (K) triglycerides (TG), (L) low density lipoprotein (LDL) and (M) high density lipoprotein (HDL). For statistical differences, * for P<0.05, while ** for P<0.01, *** for P<0.001 and **** for P<0.0001.

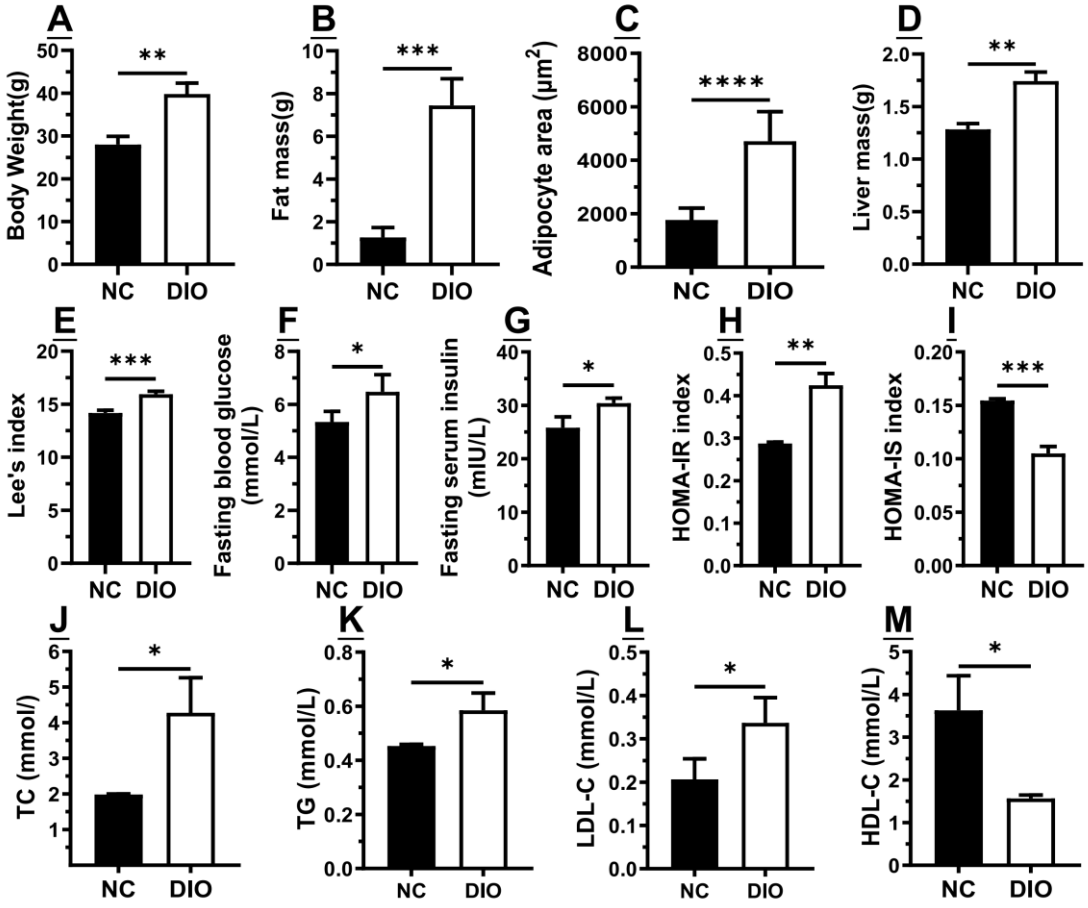

**Figure S3 High-fat diet induced the microbiota dysbiosis (n = 3).**

(A) Shannon index and (B) Chao1 index on OUT level of fecal microbiota of NC and DIO mice; (C) Phylum-level and (D) Genus-level distribution of fecal microbiota of NC and DIO mice; (E) Principal components analysis (PCA) of fecal microbiota. For statistical differences, ns indicates P>0.05, while * for P<0.05.

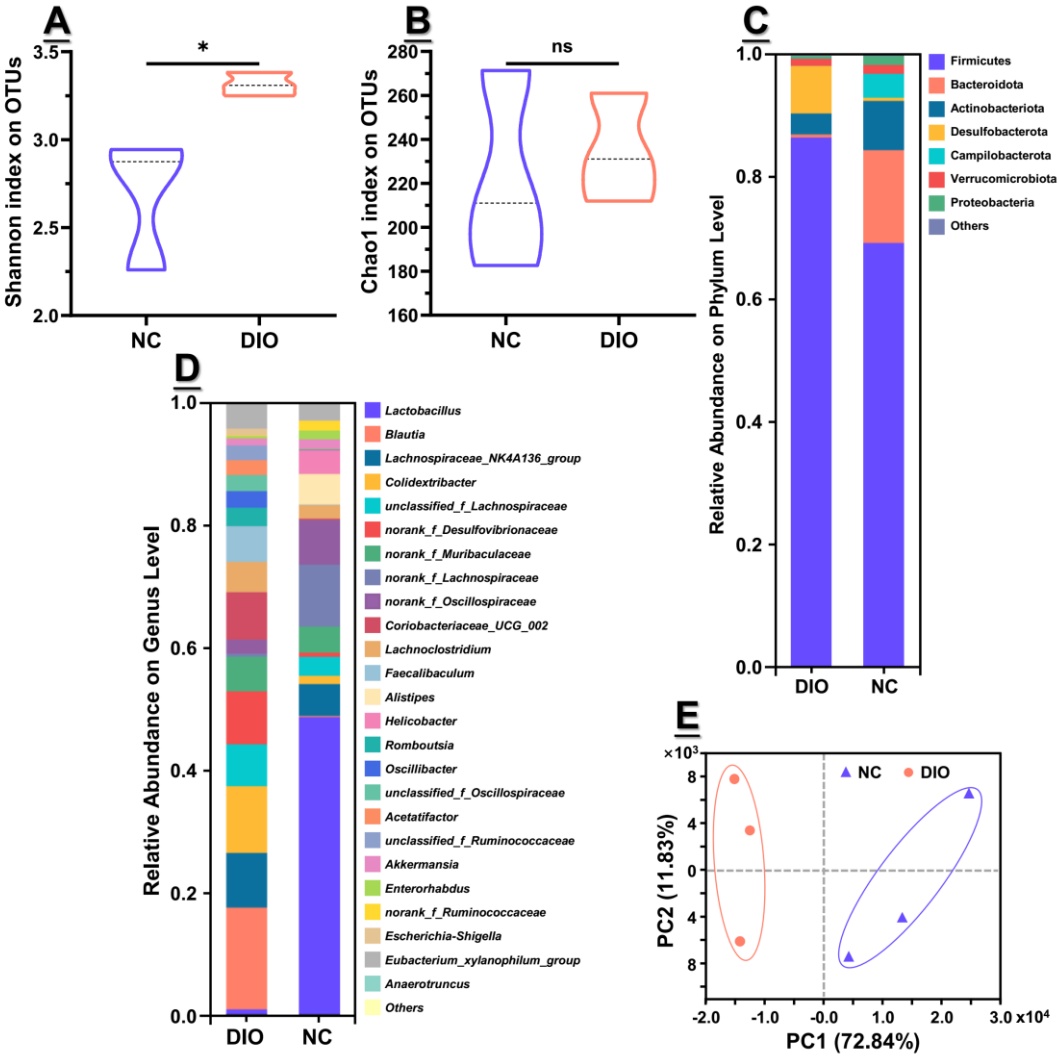

**Figure S4 Obesity was intensified on pseudo-sterile mice receiving feces of DIO mice after fed with high-fat diet.**

(A) habitus of mice and (B) H&E staining of their epididymis fat.