

Targeted delivery of butyrate improves glucose homeostasis, reduces hepatic lipid accumulation and inflammation in db/db mice

Signe Schultz Pedersen, Michala Prause, Christina Sørensen, Joachim Størling, Thomas Moritz, Eliana Mariño and Nils Billestrup

Supplementary Materials

List of materials included:

Figures:

Figure S1. Baseline characteristics of db/db mice.

Figure S2. Cytokines in plasma.

Tables:

Table S1. Diet formulations.

Table S2. Primers used for RT-qPCR on liver and fat tissue.

Table S3. Primers used for RT-qPCR on islets.

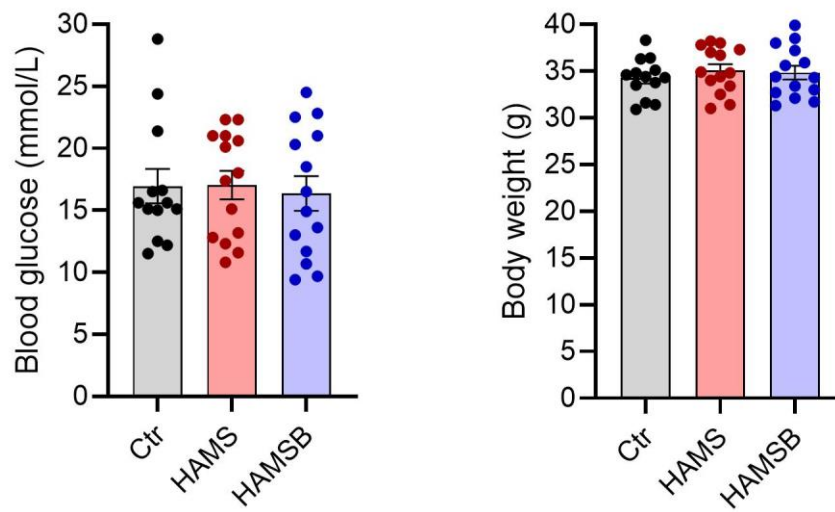


Figure S1. Baseline characteristics of db/db mice. Blood glucose (fed) and body weight were measured before initiation of the diet intervention. Bars show mean \pm SEM for the three different diet groups: Ctr, HAMS and HAMSB. Ctr, Control; HAMS, high-amylose maize starch; HAMSB, butyrylated HAMS

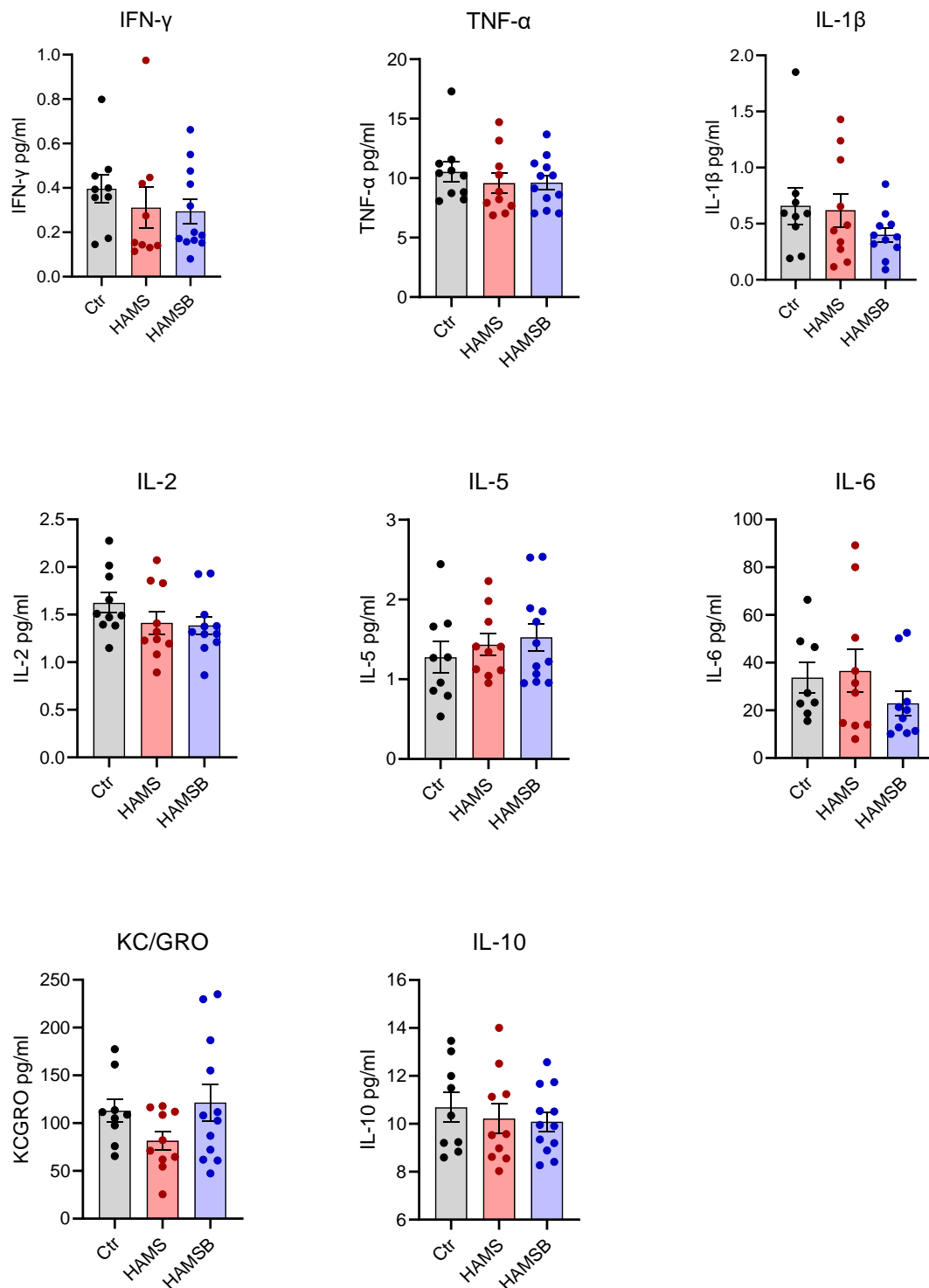


Figure S2. Cytokines in plasma. Db/db mice were fed three different diets: Ctr, HAMS or HAMSB. After 5 weeks on diet, plasma was collected for analysis of interferon gamma (IFN- γ), tumor necrosis factor alpha (TNF- α), interleukin-1 beta (IL-1 β), interleukin-2 (IL-2), interleukin-5 (IL-5), interleukin-6 (IL-6), keratinocyte chemoattractant/human growth-regulated oncogene (KC/GRO) and interleukin-10 (IL-10). Bars show mean \pm SEM.

Table S1. Diet formulations.

Numbers indicate g/kg

| | Ctr | HAMS | HAMSB |
|--|------------|-------------|--------------|
| HAMS | - | 150 | - |
| HAMSB | - | - | 150 |
| Casein | 200 | 200 | 200 |
| Sucrose | 100 | 100 | 100 |
| Corn starch | 397.5 | 247.5 | 247.5 |
| Maltodextrin | 132 | 132 | 132 |
| Soybean oil | 70 | 70 | 70 |
| Cellulose | 50 | 50 | 50 |
| Mineral Mix, AIN-93G-MX (94046) | 35 | 35 | 35 |
| Vitamin Mix, AIN-93-VX (94047) | 10 | 10 | 10 |
| L-cystine | 3 | 3 | 3 |
| Choline Bitartrate | 2.5 | 2.5 | 2.5 |

| | % by weight | % kcal from |
|---------------------|--------------------|--------------------|
| Protein | 17.7 | 18.8 |
| Carbohydrate | 60.1 | 63.9 |
| Fat | 7.2 | 17.2 |
| kcal/g | 3.8 | |

Table S2. Primers used for RT-qPCR on liver and fat tissue.

| | Forward primer | Reverse primer |
|----------------|------------------------------|------------------------------|
| <i>Fasn</i> | 5' TCCAGTTAGAGCAGGACAAGC 3' | 5' GGAGGAGACCCAGCGGAT 3' |
| <i>Scd1</i> | 5' ACATGCTGTTTTGATGCTAGGG 3' | 5' GAAGAAACAAAGCTGGACACCA 3' |
| <i>Srebp1c</i> | 5' AGACCCTGGTGAGTGGAGG 3' | 5' TGGATGGGCAGTTTGTCTGT 3' |
| <i>Acaca</i> | 5' AGGCACTCTGATCTGGTCAC 3' | 5' ATAGCTTCCAGGAGCAGTCG 3' |
| <i>Cd36</i> | 5' GATGTGCAAAACCCAGATGA 3' | 5' GATGTGCAAAACCCAGATGA 3' |
| <i>Cpt1a</i> | 5' GGCATAAACGCAGAGCATTCTG 3' | 5' CAGTGTCCATCCTCTGAGTAGC 3' |
| <i>Pck1</i> | 5' ATGAAAGGCCGACCATGTA 3' | 5' GCACAGATATGCCCATCCGA 3' |
| <i>G6pase</i> | 5' TGGTAGCCCTGTCTTTCTTTG 3' | 5' TTCCAGCATTCACACTTTCCT 3' |
| <i>Cd68</i> | 5' ATGCCACAGTTTCTCCCACC 3' | 5' ACGTGTAGTTCCCAAGAGCC 3' |
| <i>F4/80</i> | 5' ACTTTCAAGGCCCAGGAGTG 3' | 5' TGCAGACTGAGTTAGGACCAC 3' |
| <i>TNF-α</i> | 5' TGTACCTTGTCTACTCCCAGGT 3' | 5' GCAGAGAGGAGGTTGACTTTC 3' |
| <i>Ppia</i> | 5' GAGCTGTTTGCAGACAAAGTTC 3' | 5' CCCTGGCACATGAATCCTGG 3' |

Table S3. Primers used for RT-qPCR on islets.

| | |
|-------------|---------------|
| <i>Ppia</i> | Rn00690933_m1 |
| <i>Ucn3</i> | Mm00453206_s1 |
| <i>Pdx1</i> | Mm00435565_m1 |
| <i>MafA</i> | Rn00845206_s1 |
| <i>Ins1</i> | Mm01950294_s1 |
| <i>Ins2</i> | Mm00731595_gh |