

## Supplemental Tables

**Table S1.** Sample Metadata

| Animal ID | Breeding Round | PD   | Mother ID | Treatment | Litter ID | Sex    | NCBI Accession |
|-----------|----------------|------|-----------|-----------|-----------|--------|----------------|
| Dam-011   | 1              | N/A  | N/A       | Control   | CONT-L1   | FEMALE | SAMN14007843   |
| Dam-011   | 2              | N/A  | N/A       | MSEW      | MSEW-L4   | FEMALE | SAMN14007845   |
| Dam-012   | 1              | N/A  | N/A       | Control   | CONT-L2   | FEMALE | SAMN14007846   |
| Dam-012   | 2              | N/A  | N/A       | MSEW      | MSEW-L5   | FEMALE | SAMN14007848   |
| Dam-013   | 1              | N/A  | N/A       | Control   | CONT-L3   | FEMALE | SAMN14007849   |
| Dam-013   | 2              | N/A  | N/A       | MSEW      | MSEW-L6   | FEMALE | SAMN14007851   |
| Dam-014   | 1              | N/A  | N/A       | MSEW      | MSEW-L1   | FEMALE | SAMN14007852   |
| Dam-015   | 1              | N/A  | N/A       | MSEW      | MSEW-L3   | FEMALE | SAMN14007853   |
| Dam-015   | 2              | N/A  | N/A       | Control   | CONT-L4   | FEMALE | SAMN14007854   |
| Dam-016   | 1              | N/A  | N/A       | MSEW      | MSEW-L2   | FEMALE | SAMN14007856   |
| Dam-016   | 2              | N/A  | N/A       | Control   | CONT-L5   | FEMALE | SAMN14007857   |
| C-L1-01   | 1              | PD2  | 11        | Control   | CONT-L1   | MALE   | SAMN14007774   |
| C-L1-02   | 1              | PD2  | 11        | Control   | CONT-L1   | FEMALE | SAMN14007775   |
| C-L1-03   | 1              | PD10 | 11        | Control   | CONT-L1   | MALE   | SAMN14007776   |
| C-L1-05   | 1              | PD28 | 11        | Control   | CONT-L1   | MALE   | SAMN14007777   |
| C-L1-06   | 1              | PD28 | 11        | Control   | CONT-L1   | MALE   | SAMN14007778   |
| C-L2-01   | 1              | PD2  | 12        | Control   | CONT-L2   | MALE   | SAMN14007779   |
| C-L2-02   | 1              | PD2  | 12        | Control   | CONT-L2   | FEMALE | SAMN14007780   |
| C-L2-03   | 1              | PD10 | 12        | Control   | CONT-L2   | MALE   | SAMN14007781   |
| C-L2-04   | 1              | PD10 | 12        | Control   | CONT-L2   | FEMALE | SAMN14007782   |
| C-L2-05   | 1              | PD10 | 12        | Control   | CONT-L2   | FEMALE | SAMN14007783   |
| C-L2-06   | 1              | PD28 | 12        | Control   | CONT-L2   | FEMALE | SAMN14007784   |
| C-L2-07   | 1              | PD28 | 12        | Control   | CONT-L2   | FEMALE | SAMN14007785   |
| C-L2-08   | 1              | PD28 | 12        | Control   | CONT-L2   | MALE   | SAMN14007786   |
| C-L3-01   | 1              | PD2  | 13        | Control   | CONT-L3   | MALE   | SAMN14007787   |
| C-L3-02   | 1              | PD2  | 13        | Control   | CONT-L3   | FEMALE | SAMN14007788   |
| C-L3-04   | 1              | PD10 | 13        | Control   | CONT-L3   | MALE   | SAMN14007789   |
| C-L3-05   | 1              | PD28 | 13        | Control   | CONT-L3   | MALE   | SAMN14007790   |
| C-L3-06   | 1              | PD28 | 13        | Control   | CONT-L3   | MALE   | SAMN14007791   |
| C-L3-07   | 1              | PD28 | 13        | Control   | CONT-L3   | MALE   | SAMN14007792   |
| C-L4-01   | 2              | PD2  | 15        | Control   | CONT-L4   | MALE   | SAMN14007793   |
| C-L4-02   | 2              | PD2  | 15        | Control   | CONT-L4   | FEMALE | SAMN14007794   |
| C-L4-03   | 2              | PD10 | 15        | Control   | CONT-L4   | MALE   | SAMN14007795   |

|         |   |      |    |         |         |        |              |
|---------|---|------|----|---------|---------|--------|--------------|
| C-L4-04 | 2 | PD28 | 15 | Control | CONT-L4 | MALE   | SAMN14007796 |
| C-L4-05 | 2 | PD28 | 15 | Control | CONT-L4 | MALE   | SAMN14007797 |
| C-L5-01 | 2 | PD2  | 16 | Control | CONT-L5 | MALE   | SAMN14007798 |
| C-L5-02 | 2 | PD2  | 16 | Control | CONT-L5 | FEMALE | SAMN14007799 |
| C-L5-03 | 2 | PD10 | 16 | Control | CONT-L5 | MALE   | SAMN14007800 |
| C-L5-04 | 2 | PD10 | 16 | Control | CONT-L5 | FEMALE | SAMN14007801 |
| C-L5-05 | 2 | PD10 | 16 | Control | CONT-L5 | MALE   | SAMN14007802 |
| C-L5-06 | 2 | PD28 | 16 | Control | CONT-L5 | FEMALE | SAMN14007803 |
| C-L5-07 | 2 | PD28 | 16 | Control | CONT-L5 | MALE   | SAMN14007804 |
| C-L5-08 | 2 | PD28 | 16 | Control | CONT-L5 | FEMALE | SAMN14007805 |
| M-L1-01 | 1 | PD2  | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007806 |
| M-L1-03 | 1 | PD10 | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007807 |
| M-L1-04 | 1 | PD10 | 14 | MSEW    | MSEW-L1 | FEMALE | SAMN14007808 |
| M-L1-05 | 1 | PD10 | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007809 |
| M-L1-06 | 1 | PD10 | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007810 |
| M-L1-08 | 1 | PD28 | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007811 |
| M-L1-09 | 1 | PD28 | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007812 |
| M-L1-10 | 1 | PD28 | 14 | MSEW    | MSEW-L1 | FEMALE | SAMN14007813 |
| M-L1-11 | 1 | PD28 | 14 | MSEW    | MSEW-L1 | FEMALE | SAMN14007814 |
| M-L1-12 | 1 | PD28 | 14 | MSEW    | MSEW-L1 | MALE   | SAMN14007815 |
| M-L2-01 | 1 | PD2  | 16 | MSEW    | MSEW-L2 | MALE   | SAMN14007816 |
| M-L2-02 | 1 | PD2  | 16 | MSEW    | MSEW-L2 | FEMALE | SAMN14007817 |
| M-L2-03 | 1 | PD10 | 16 | MSEW    | MSEW-L2 | MALE   | SAMN14007818 |
| M-L2-04 | 1 | PD10 | 16 | MSEW    | MSEW-L2 | FEMALE | SAMN14007819 |
| M-L2-05 | 1 | PD10 | 16 | MSEW    | MSEW-L2 | MALE   | SAMN14007820 |
| M-L2-06 | 1 | PD10 | 16 | MSEW    | MSEW-L2 | FEMALE | SAMN14007821 |
| M-L2-07 | 1 | PD28 | 16 | MSEW    | MSEW-L2 | FEMALE | SAMN14007822 |
| M-L2-08 | 1 | PD28 | 16 | MSEW    | MSEW-L2 | MALE   | SAMN14007823 |
| M-L2-09 | 1 | PD28 | 16 | MSEW    | MSEW-L2 | MALE   | SAMN14007824 |
| M-L3-01 | 1 | PD2  | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007825 |
| M-L3-02 | 1 | PD2  | 15 | MSEW    | MSEW-L3 | FEMALE | SAMN14007826 |
| M-L3-03 | 1 | PD10 | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007827 |
| M-L3-04 | 1 | PD10 | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007828 |
| M-L3-05 | 1 | PD10 | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007829 |
| M-L3-06 | 1 | PD10 | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007830 |
| M-L3-07 | 1 | PD28 | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007831 |
| M-L3-08 | 1 | PD28 | 15 | MSEW    | MSEW-L3 | FEMALE | SAMN14007832 |
| M-L3-09 | 1 | PD28 | 15 | MSEW    | MSEW-L3 | MALE   | SAMN14007833 |

|         |   |      |    |      |         |        |              |
|---------|---|------|----|------|---------|--------|--------------|
| M-L3-10 | 1 | PD28 | 15 | MSEW | MSEW-L3 | MALE   | SAMN14007834 |
| M-L4-03 | 2 | PD10 | 11 | MSEW | MSEW-L4 | FEMALE | SAMN14007835 |
| M-L5-01 | 2 | PD2  | 12 | MSEW | MSEW-L5 | MALE   | SAMN14007836 |
| M-L5-02 | 2 | PD2  | 12 | MSEW | MSEW-L5 | FEMALE | SAMN14007837 |
| M-L5-03 | 2 | PD10 | 12 | MSEW | MSEW-L5 | MALE   | SAMN14007838 |
| M-L5-04 | 2 | PD10 | 12 | MSEW | MSEW-L5 | MALE   | SAMN14007839 |
| M-L5-05 | 2 | PD10 | 12 | MSEW | MSEW-L5 | FEMALE | SAMN14007840 |
| M-L6-01 | 2 | PD2  | 13 | MSEW | MSEW-L6 | MALE   | SAMN14007841 |
| M-L6-02 | 2 | PD2  | 13 | MSEW | MSEW-L6 | MALE   | SAMN14007842 |

**Table S2.** Analysis of variance table for the effect of treatment on body weight using the generalized linear mixed model (GLMM) framework. The model formula in the R language was: `lmer(body weight ~ treatment + (1|dam identity))`. Denominator degrees of freedom and *P*-values for the F test were approximated with the `lmerTest` package.

| PD2              |       |       |       |       |       |        |
|------------------|-------|-------|-------|-------|-------|--------|
|                  | SS    | MS    | NumDf | DenDf | F     | Pr(>F) |
| <i>treatment</i> | 0.004 | 0.004 | 1     | 1.836 | 0.438 | 0.581  |

| PD5              |       |       |       |       |       |        |
|------------------|-------|-------|-------|-------|-------|--------|
|                  | SS    | MS    | NumDf | DenDf | F     | Pr(>F) |
| <i>treatment</i> | 0.002 | 0.002 | 1     | 2.665 | 0.073 | 0.807  |

| PD11             |      |      |       |       |       |        |
|------------------|------|------|-------|-------|-------|--------|
|                  | SS   | MS   | NumDf | DenDf | F     | Pr(>F) |
| <i>treatment</i> | 0.01 | 0.01 | 1     | 2.823 | 0.068 | 0.813  |

| PD15             |       |       |       |       |       |        |
|------------------|-------|-------|-------|-------|-------|--------|
|                  | SS    | MS    | NumDf | DenDf | F     | Pr(>F) |
| <i>treatment</i> | 0.068 | 0.068 | 1     | 2.931 | 0.277 | 0.636  |

Abbreviations: PD, post-natal day; SS, sum of squares; MS, mean squares; NumDf, numerator degrees of freedom; DenDf, denominator degrees of freedom; F, F-test; R<sup>2</sup>, R-squared; Pr(>F), *P*-value associated with the F-statistic

**Table S3.** Analysis of composition of microbiomes (ANCOM) results and taxonomic classification for differentially abundant ASVs at PD10. The ANCOM test statistic  $W$  represents the number of times the ratio between the given taxon and other taxa was significantly different between the two treatment groups.  $W = 77$  means the ratios of the given taxon and 77 other taxa were detected to be significantly different between the two treatment groups. The threshold for significance was set to a Benjamini-Hochberg false discovery rate corrected  $P$ -value  $< 0.05$ . Significance at threshold 0.7 corresponds to the ratio being significantly different between treatment groups for 70% of the samples.

| Taxa id | $W$<br>statistic | Detected<br>0.9 | Detected<br>0.8 | Detected<br>0.7 | Taxonomy   |
|---------|------------------|-----------------|-----------------|-----------------|--|
| ASV22   | 77               | TRUE            | TRUE            | TRUE            | Firmicutes, Bacilli, Lactobacillales,<br>Enterococcaceae, Enterococcus |

Abbreviations: ASV, amplicon sequence variation;  $W$  statistic, ANCOM test statistic

**Table S4.** Analysis of composition of microbiomes (ANCOM) results and taxonomic classification for differentially abundant ASVs at PD28. The ANCOM test statistic  $W$  represents the number of times the ratio between the given taxon and other taxa was significantly different between the two treatment groups.  $W = 288$  means the ratios of the given taxon and 288 other taxa were detected to be significantly different between the two treatment groups. The threshold for significance was set to a Benjamini-Hochberg false discovery rate corrected  $P$ -value  $< 0.05$ . Significance at threshold 0.7 corresponds to the ratio being significantly different between treatment groups for 70% of the samples.

| Taxa id | W statistic | Detected 0.9 | Detected 0.8 | Detected 0.7 | Taxonomy  |
|---------|-------------|--------------|--------------|--------------|---|
| ASV42   | 288         | TRUE         | TRUE         | TRUE         | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Incertae Sedis                          |
| ASV82   | 285         | TRUE         | TRUE         | TRUE         | Firmicutes, Clostridia, Clostridiales, Clostridiaceae, Clostridium sensu stricto 1                |
| ASV20   | 284         | TRUE         | TRUE         | TRUE         | Firmicutes, Clostridia, Clostridiales, Clostridiaceae, Clostridium sensu stricto 1                |
| ASV34   | 280         | TRUE         | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-001                  |
| ASV277  | 270         | TRUE         | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnoclostridium                        |
| ASV9    | 268         | TRUE         | TRUE         | TRUE         | Bacteroidota, Bacteroidia, Bacteroidales, Muribaculaceae, Muribaculaceae                          |
| ASV64   | 268         | TRUE         | TRUE         | TRUE         | Firmicutes, Clostridia, Clostridia vadinBB60 group, Clostridia vadinBB60 group                    |
| ASV33   | 263         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-001                  |
| ASV119  | 263         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Marvinbryantia                           |
| ASV197  | 262         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, UCG-005,                               |
| ASV90   | 258         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Clostridia UCG-014, Clostridia UCG-014, Clostridia UCG-014                |
| ASV130  | 254         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Marvinbryantia                           |
| ASV110  | 249         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnoclostridium                        |
| ASV117  | 247         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnoclostridium                        |
| ASV76   | 246         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnoclostridium                        |
| ASV13   | 244         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-001                  |
| ASV59   | 242         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae NK4A136 group, bacterium |
| ASV276  | 236         | FALSE        | TRUE         | TRUE         | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Ruminococcaceae                         |
| ASV315  | 233         | FALSE        | FALSE        | TRUE         | Firmicutes, Clostridia, Clostridia vadinBB60 group, Clostridia vadinBB60 group                    |

|        |     |       |       |      |  |
|--------|-----|-------|-------|------|--|
| ASV208 | 232 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Clostridia vadinBB60 group, Clostridia vadinBB60 group |
| ASV195 | 231 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Tyzzerella            |
| ASV295 | 231 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Clostridia vadinBB60 group, Clostridia vadinBB60 group |
| ASV98  | 228 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Clostridia UCG-014, Clostridia UCG-014                 |
| ASV41  | 227 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, [Eubacterium]         |
| ASV149 | 226 | FALSE | FALSE | TRUE | xylanophilum group   |
| ASV203 | 224 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Marvinbryantia        |
| ASV261 | 223 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Oscillibacter       |
| ASV107 | 219 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae                        |
| ASV108 | 219 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae                        |
| ASV67  | 211 | FALSE | FALSE | TRUE | Bacteroidota, Bacteroidia, Bacteroidales, Muribaculaceae                       |
| ASV352 | 208 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Intestinimonas      |
| ASV137 | 206 | FALSE | FALSE | TRUE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, [Eubacterium]         |
|        |     |       |       |      | xylanophilum group   |

Abbreviations: ASV, amplicon sequence variation;  $W$  statistic, ANCOM test statistic

**Table S5.** Analysis of composition of microbiomes (ANCOM) results and taxonomic classification for differentially abundant genera at PD10. The ANCOM test statistic  $W$  represents the number of times the ratio between the given taxon and other taxa was significantly different between the two treatment groups.  $W = 54$  means the ratios of the given taxon and 54 other taxa were detected to be significantly different between the two treatment groups. The threshold for significance was set to a Benjamini-Hochberg false discovery rate corrected  $P$ -value  $< 0.05$ . Significance at threshold 0.7 corresponds to the ratio being significantly different between treatment groups for 70% of the samples.

| $W$ | Detected<br>0.9 | Detected<br>0.8 | Detected<br>0.7 | Taxa   |
|-----|-----------------|-----------------|-----------------|--|
| 54  | TRUE            | TRUE            | TRUE            | Firmicutes, Bacilli, Lactobacillales, Enterococcaceae, Enterococcus                      |
| 15  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Incertae Sedis                 |
| 2   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Enterobacterales, Enterobacteriaceae, Plesiomonas   |
| 2   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Flavobacteriales, Weeksellaceae, Cloacibacterium              |
| 1   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Erysipelotrichales, Erysipelotrichaceae, Ileibacterium              |
| 1   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, A2                              |
| 1   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, ASF356                          |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Lactobacillales, Lactobacillaceae, Lactobacillus                    |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Staphylococcales, Staphylococcaceae, Staphylococcus                 |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae NK4A136 group   |
| 0   | FALSE           | FALSE           | FALSE           | Actinobacteriota, Actinobacteria, Bifidobacteriales, Bifidobacteriaceae, Bifidobacterium |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Clostridiales, Clostridiaceae, Clostridium sensu stricto 1       |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-001         |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Roseburia                       |
| 0   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Bacteroidaceae, Bacteroides                    |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae FCS020 group    |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Intestinimonas                |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Oscillibacter                 |
| 0   | FALSE           | FALSE           | FALSE           | Verrucomicrobiota, Verrucomicrobiae, Verrucomicrobiales, Akkermansiaceae, Akkermansia    |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Achleplasmatales, Achleplasmataceae, Anaeroplasma                   |



|   |       |       |       |   |
|---|-------|-------|-------|---|
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, [Eubacterium] xylanophilum group           |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Burkholderiales, Sutterellaceae, Parasutterella                |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Colidextribacter                         |
| 0 | FALSE | FALSE | FALSE | Actinobacteriota, Coriobacteriia, Coriobacteriales, Eggerthellaceae, Parvibacter                    |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Erysipelotrichales, Erysipelotrichaceae, Turicibacter                          |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Butyricicoccaceae, UCG-009                                 |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Acetatifactor                              |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, [Eubacterium] siraeum group               |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-006                    |
| 0 | FALSE | FALSE | FALSE | Actinobacteriota, Coriobacteriia, Coriobacteriales, Atopobiaceae, Coriobacteriaceae UCG-002         |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Marvinbryantia                             |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Erysipelotrichales, Erysipelatoclostridiaceae, Erysipelatoclostridium          |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Pseudomonadales, Pseudomonadaceae, Pseudomonas                 |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Ruminococcus                              |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, GCA-900066575                              |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Aeromonadales, Aeromonadaceae, Aeromonas                       |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnoclostridium                          |
| 0 | FALSE | FALSE | FALSE | Actinobacteriota, Coriobacteriia, Coriobacteriales, Eggerthellaceae, Enterorhabdus                  |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Tuzzerella                                 |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-004                    |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Tyzzerella                                 |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, UCG-005                                  |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, [Eubacterium] ventriosum group             |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Peptostreptococcaceae, Romboutsia      |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Peptostreptococcaceae, Paraclostridium |

|   |       |       |       |  |
|---|-------|-------|-------|--|
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Anaerovoracaceae, [Eubacterium] nodatum group |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Pseudomonadales, Moraxellaceae, Acinetobacter                         |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Burkholderiales, Comamonadaceae, Delftia                              |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Paenibacillales, Paenibacillaceae, Paenibacillus                                      |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Burkholderiales, Comamonadaceae, Ottowia                              |
| 0 | FALSE | FALSE | FALSE | Actinobacteriota, Actinobacteria, Streptomycetales, Streptomyces   |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Enterobacterales, Enterobacteriaceae, Klebsiella                      |
| 0 | FALSE | FALSE | FALSE | Bacteroidota, Bacteroidia, Bacteroidales, Tannerellaceae, Macellibacteroides                               |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Bacillales, Planococcaceae, Lysinibacillus  |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Enterobacterales, Enterobacteriaceae, Escherichia-Shigella            |

Abbreviations: ASV, amplicon sequence variation; *W*, ANCOM test statistic

**Table S6.** Analysis of composition of microbiomes (ANCOM) results and taxonomic classification for differentially abundant families at PD10. The ANCOM test statistic  $W$  represents the number of times the ratio between the given taxon and other taxa was significantly different between the two treatment groups.  $W = 30$  means the ratios of the given taxon and 30 other taxa were detected to be significantly different between the two treatment groups. The threshold for significance was set to a Benjamini-Hochberg false discovery rate corrected  $P$ -value  $< 0.05$ . Significance at threshold 0.7 corresponds to the ratio being significantly different between treatment groups for 70% of the samples.

| $W$ | Detected<br>0.9 | Detected<br>0.8 | Detected<br>0.7 | Taxa   |
|-----|-----------------|-----------------|-----------------|--|
| 30  | TRUE            | TRUE            | TRUE            | Firmicutes, Bacilli, Lactobacillales, Enterococcaceae                          |
| 17  | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Burkholderiales, Comamonadaceae           |
| 13  | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Erysipelotrichales, Erysipelotrichaceae                   |
| 6   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae                       |
| 5   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Acholeplasmatales, Acholeplasmataceae                     |
| 5   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Flavobacteriales, Weeksellaceae                     |
| 3   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Burkholderiales, Sutterellaceae           |
| 3   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Anaerovoracaceae  |
| 2   | FALSE           | FALSE           | FALSE           | Actinobacteriota, Actinobacteria, Bifidobacteriales, Bifidobacteriaceae        |
| 2   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, [Eubacterium] coprostanoligenes group |
| 2   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Enterobacteriales, Enterobacteriaceae     |
| 1   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Butyricicoccaceae                     |
| 1   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Tannerellaceae                       |
| 1   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Bacillales, Planococcaceae                                |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Lactobacillales, Lactobacillaceae                         |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Staphylococcales, Staphylococcaceae                       |
| 0   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Muribaculaceae                       |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Clostridiales, Clostridiaceae                          |
| 0   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Bacteroidaceae                       |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae                      |
| 0   | FALSE           | FALSE           | FALSE           | Verrucomicrobiota, Verrucomicrobiae, Verrucomicrobiales, Akkermansiaceae       |
| 0   | FALSE           | FALSE           | FALSE           | Actinobacteriota, Coriobacteriia, Coriobacteriales, Eggerthellaceae            |
| 0   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Peptococcales, Peptococcaceae                          |
| 0   | FALSE           | FALSE           | FALSE           | Actinobacteriota, Coriobacteriia, Coriobacteriales, Atopobiaceae               |

|   |       |       |       |   |
|---|-------|-------|-------|---|
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Erysipelotrichales,<br>Erysipelatoclostridiaceae                 |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Pseudomonadales,<br>Pseudomonadaceae             |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Aeromonadales,<br>Aeromonadaceae                 |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae                               |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales,<br>Peptostreptococcaceae |
| 0 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Pseudomonadales,<br>Moraxellaceae                |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Paenibacillales, Paenibacillaceae                                |
| 0 | FALSE | FALSE | FALSE | Actinobacteriota, Actinobacteria, Streptomycetales,<br>Streptomycetaceae              |

Abbreviations: ASV, amplicon sequence variation;  $W$ , ANCOM test statistic

**Table S7.** Analysis of composition of microbiomes (ANCOM) results and taxonomic classification for differentially abundant genera at PD28. The ANCOM test statistic  $W$  represents the number of times the ratio between the given taxon and other taxa was significantly different between the two treatment groups.  $W = 53$  means the ratios of the given taxon and 53 other taxa were detected to be significantly different between the two treatment groups. The threshold for significance was set to a Benjamini-Hochberg false discovery rate corrected  $P$ -value  $< 0.05$ . Significance at threshold 0.7 corresponds to the ratio being significantly different between treatment groups for 70% of the samples.

| $W$ | Detected<br>0.9 | Detected<br>0.8 | Detected<br>0.7 | Taxa   |
|-----|-----------------|-----------------|-----------------|--|
| 53  | FALSE           | TRUE            | TRUE            | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Tyzzerella  |
| 47  | FALSE           | FALSE           | TRUE            | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Ruminococcus                                     |
| 47  | FALSE           | FALSE           | TRUE            | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, UCG-005   |
| 37  | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Lactobacillales, Lactobacillaceae, Lactobacillus                                      |
| 37  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Peptostreptococcaceae, Romboutsia             |
| 30  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Marvinbryantia                                    |
| 28  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Clostridiales, Clostridiaceae, Clostridium sensu stricto 1                         |
| 23  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Anaerovoracaceae, [Eubacterium] nodatum group |
| 20  | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Bacteroidaceae, Bacteroides                                      |
| 19  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Oscillibacter                                   |
| 15  | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Pseudomonadales, Moraxellaceae, Acinetobacter                         |
| 11  | FALSE           | FALSE           | FALSE           | Actinobacteriota, Coriobacteriia, Coriobacteriales, Eggerthellaceae, Enterorhabdus                         |
| 10  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, GCA-900066575                                     |
| 10  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, NK4A214 group                                   |
| 8   | FALSE           | FALSE           | FALSE           | Verrucomicrobiota, Verrucomicrobiae, Verrucomicrobiales, Akkermansiaceae, Akkermansia                      |
| 8   | FALSE           | FALSE           | FALSE           | Actinobacteriota, Coriobacteriia, Coriobacteriales, Eggerthellaceae, Parvibacter                           |
| 8   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Paludicola                                       |
| 6   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Erysipelotrichales, Erysipelotrichaceae, Turicibacter                                 |
| 6   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Burkholderiales, Sutterellaceae, Parasutterella                       |

|   |       |       |       |   |
|---|-------|-------|-------|---|
| 6 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnoclostridium                                |
| 6 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Pseudomonadales, Pseudomonadaceae, Pseudomonas                       |
| 5 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Roseburia  |
| 5 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Acholeplasmatales, Acholeplasmataceae, Anaeroplasma                                  |
| 5 | FALSE | FALSE | FALSE | Actinobacteriota, Coriobacteriia, Coriobacteriales, Atopobiaceae, Coriobacteriaceae UCG-002               |
| 5 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Monoglobales, Monoglobaceae, Monoglobus   |
| 4 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Erysipelotrichales, Erysipelatoclostridiaceae, Erysipelatoclostridium                |
| 4 | FALSE | FALSE | FALSE | Actinobacteriota, Coriobacteriia, Coriobacteriales, Eggerthellaceae, Adlercreutzia                        |
| 4 | FALSE | FALSE | FALSE | Bacteroidota, Bacteroidia, Flavobacteriales, Weeksellaceae, Cloacibacterium                               |
| 4 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, UCG-003  |
| 3 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Staphylococcales, Staphylococcaceae, Staphylococcus                                  |
| 3 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-001                          |
| 3 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae FCS020 group                     |
| 3 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Colidextribacter                               |
| 3 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, [Eubacterium] siraeum group                     |
| 3 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Burkholderiales, Comamonadaceae, Ottowia                             |
| 2 | FALSE | FALSE | FALSE | Actinobacteriota, Actinobacteria, Bifidobacteriales, Bifidobacteriaceae, Bifidobacterium                  |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Lactobacillales, Enterococcaceae, Enterococcus                                       |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Butyricicoccaceae, UCG-009                                       |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Acetatifactor                                    |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Tuzzerella                                       |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae AC2044 group                     |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Peptostreptococcaceae, Paraclostridium       |
| 2 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Anaerovoracaceae, [Eubacterium] brachy group |
| 1 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Incertae Sedis                                  |

|   |       |       |       |   |
|---|-------|-------|-------|---|
| 1 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Aeromonadales, Aeromonadaceae, Aeromonas,            |
| 1 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Butyricicoccaceae, Butyricoccus                  |
| 1 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Burkholderiales, Aquaspirillaceae, Microvirgula      |
| 1 | FALSE | FALSE | FALSE | Bacteroidota, Bacteroidia, Bacteroidales, Tannerellaceae, Macellibacteroides              |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae NK4A136 group    |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Erysipelotrichales, Erysipelotrichaceae, Ileibacterium               |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae, Intestinimonas                 |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, A2                               |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, [Eubacterium] xylanophilum group |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-006          |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, ASF356                           |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Lachnospiraceae UCG-004          |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, [Eubacterium] ventriosum group   |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae, Dorea                            |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Anaerotruncus                   |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae, Harryflintia                    |

Abbreviations: ASV, amplicon sequence variation; *W*, ANCOM test statistic

**Table S8.** Analysis of composition of microbiomes (ANCOM) results and taxonomic classification for differentially abundant families at PD28. The ANCOM test statistic  $W$  represents the number of times the ratio between the given taxon and other taxa was significantly different between the two treatment groups.  $W = 28$  means the ratios of the given taxon and 28 other taxa were detected to be significantly different between the two treatment groups. The threshold for significance was set to a Benjamini-Hochberg false discovery rate corrected  $P$ -value  $< 0.05$ . Significance at threshold 0.7 corresponds to the ratio being significantly different between treatment groups for 70% of the samples.

| $W$ | Detected<br>0.9 | Detected<br>0.8 | Detected<br>0.7 | Taxa   |
|-----|-----------------|-----------------|-----------------|--|
| 28  | TRUE            | TRUE            | TRUE            | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Peptostreptococcaceae |
| 21  | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Lactobacillales, Lactobacillaceae                             |
| 16  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Clostridiales, Clostridiaceae                              |
| 14  | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Bacteroidaceae                           |
| 14  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Peptostreptococcales-Tissierellales, Anaerovoracaceae      |
| 14  | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, UCG-010                                   |
| 12  | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Lactobacillales, Enterococcaceae                              |
| 9   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Pseudomonadales, Moraxellaceae                |
| 8   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Monoglobales, Moraxellaceae                                |
| 7   | FALSE           | FALSE           | FALSE           | Actinobacteriota, Coriobacteriia, Coriobacteriales, Moraxellaceae                  |
| 7   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Erysipelotrichales, Erysipelatoclostridiaceae                 |
| 6   | FALSE           | FALSE           | FALSE           | Verrucomicrobiota, Verrucomicrobiae, Verrucomicrobiales, Akkermansiaceae           |
| 5   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Erysipelotrichales, Erysipelotrichaceae                       |
| 4   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Flavobacteriales, Weeksellaceae                         |
| 3   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, [Eubacterium] coprostanoligenes group     |
| 2   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Muribaculaceae                           |
| 2   | FALSE           | FALSE           | FALSE           | Firmicutes, Bacilli, Acholeplasmatales, Acholeplasmataceae                         |
| 2   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Burkholderiales, Sutterellaceae               |
| 2   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Oscillospirales, Butyricicoccaceae                         |
| 2   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Pseudomonadales, Pseudomonadaceae             |
| 2   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Burkholderiales, Comamonadaceae               |
| 2   | FALSE           | FALSE           | FALSE           | Proteobacteria, Gammaproteobacteria, Enterobacteriales, Enterobacteriaceae         |
| 2   | FALSE           | FALSE           | FALSE           | Bacteroidota, Bacteroidia, Bacteroidales, Tannerellaceae                           |
| 1   | FALSE           | FALSE           | FALSE           | Firmicutes, Clostridia, Peptococcales, Peptococcaceae                              |



|   |       |       |       |  |
|---|-------|-------|-------|--|
| 1 | FALSE | FALSE | FALSE | Actinobacteriota, Coriobacteriia, Coriobacteriales,<br>Atopobiaceae        |
| 1 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Aeromonadales,<br>Aeromonadaceae      |
| 1 | FALSE | FALSE | FALSE | Proteobacteria, Gammaproteobacteria, Burkholderiales,<br>Aquaspirillaceae  |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Bacilli, Staphylococcales, Staphylococcaceae                   |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Lachnospirales, Lachnospiraceae                    |
| 0 | FALSE | FALSE | FALSE | Actinobacteriota, Actinobacteria, Bifidobacteriales,<br>Bifidobacteriaceae |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Oscillospiraceae                  |
| 0 | FALSE | FALSE | FALSE | Firmicutes, Clostridia, Oscillospirales, Ruminococcaceae                   |

Abbreviations: ASV, amplicon sequence variation;  $W$ , ANCOM test statistic

**Table S9.** Alpha diversity metrics. Numbers in () indicate SEM.

| Group                          | Replicate Number |      | Chao1 ASV Richness |         | Phylogenetic Diversity |           |
|--------------------------------|------------------|------|--------------------|---------|------------------------|-----------|
|                                | Control          | MSEW | Control            | MSEW    | Control                | MSEW      |
| PD2                            | 9                | 9    | 70(12)             | 55(11)  | 9.7(1.7)*              | 8.0(1.5)* |
| PD10                           | 9                | 16   | 43(7)*             | 37(7)*  | 5.7(0.4)               | 5.5(0.3)  |
| PD28                           | 13               | 12   | 198(8)             | 175(8)  | 20.3(0.3)              | 18.7(0.3) |
| Dam                            | 5                | 6    | 246(15)            | 236(14) | 20.9(0.7)              | 21.5(0.4) |
| Dam (014 removed) <sup>§</sup> | 5                | 5    | 246(15)            | 242(15) | 20.9(0.7)              | 21.6(0.4) |

Abbreviations: PD, post-natal day; ASVs, amplicon sequence variants; SEM, standard error of the mean

\* Data were log transformed to improve normality.

<sup>§</sup> Dam 014 did not produce a litter in the second breeding round and was excluded from paired t-test analyses. Excluding this sample did not produce significant changes in the treatment group means.

**Table S10.** Analysis of variance table for the effect of treatment on alpha diversity metrics of pup microbiota using the generalized linear mixed model (GLMM) framework. The model formula in the R language was: `lmer(alpha diversity metric ~ treatment + (1|dam identity))`. Denominator degrees of freedom and *P*-values for the F test were approximated with the `lmerTest` package.

| PD2                     |          |          |       |        |        |              |
|-------------------------|----------|----------|-------|--------|--------|--------------|
| Chao1                   | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)       |
| <i>treatment</i>        | 997.1    | 997.1    | 1     | 14.191 | 1.036  | 0.326        |
|                         |          |          |       |        |        |              |
| Phylogenetic Diversity* | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)       |
| <i>treatment</i>        | 0.125    | 0.125    | 1     | 13.843 | 0.591  | 0.455        |
|                         |          |          |       |        |        |              |
| PD10                    |          |          |       |        |        |              |
| Chao1*                  | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)       |
| <i>treatment</i>        | 0.126    | 0.126    | 1     | 23     | 0.7    | 0.411        |
|                         |          |          |       |        |        |              |
| Phylogenetic Diversity  | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)       |
| <i>treatment</i>        | 0.341    | 0.341    | 1     | 23     | 0.252  | 0.620        |
|                         |          |          |       |        |        |              |
| PD28                    |          |          |       |        |        |              |
| Chao1                   | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)       |
| <i>treatment</i>        | 2903.139 | 2903.139 | 1     | 20.395 | 4.969  | <b>0.037</b> |
|                         |          |          |       |        |        |              |
| Phylogenetic Diversity  | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)       |
| <i>treatment</i>        | 14.27    | 14.27    | 1     | 18.967 | 11.947 | <b>0.003</b> |

Abbreviations: PD, post-natal day; SS, sum of squares; MS, mean squares; NumDf, numerator degrees of freedom; DenDf, denominator degrees of freedom; F, F-test; R<sup>2</sup>, R-squared; Pr(>F), *P*-value associated with the F-statistic

\* Data were log transformed to improve normality before statistical analyses

**Table S11.** The effect of treatment on microbial community composition based on Aitchison distance. Significance was assessed with permutational analysis of variance (PERMANOVA) and the linear model framework stratifying permutations by *dam identity*.

| PD2              |    |            |          |       |                |        |
|------------------|----|------------|----------|-------|----------------|--------|
|                  | Df | SS         | MS       | F     | R <sup>2</sup> | Pr(>F) |
| <i>treatment</i> | 1  | 7676.435   | 7676.435 | 1.029 | 0.057          | 0.264  |
| Residuals        | 17 | 126830.070 | 7460.592 |       | 0.943          |        |
| Total            | 18 | 134506.505 |          |       | 1.000          |        |

| PD10             |    |           |          |       |                |        |
|------------------|----|-----------|----------|-------|----------------|--------|
|                  | Df | SS        | MS       | F     | R <sup>2</sup> | Pr(>F) |
| <i>treatment</i> | 1  | 3181.501  | 3181.501 | 1.522 | 0.062          | 0.0645 |
| Residuals        | 23 | 48089.248 | 2090.837 |       | 0.938          |        |
| Total            | 24 | 51270.749 |          |       | 1.000          |        |

| PD28             |    |           |          |       |                |               |
|------------------|----|-----------|----------|-------|----------------|---------------|
|                  | Df | SS        | MS       | F     | R <sup>2</sup> | Pr(>F)        |
| <i>treatment</i> | 1  | 8397.309  | 8397.309 | 2.294 | 0.091          | <b>0.0021</b> |
| Residuals        | 23 | 84185.302 | 3660.231 |       | 0.909          |               |
| Total            | 24 | 92582.612 |          |       | 1.000          |               |

| Dam              |    |           |          |       |                |        |
|------------------|----|-----------|----------|-------|----------------|--------|
|                  | Df | SS        | MS       | F     | R <sup>2</sup> | Pr(>F) |
| <i>treatment</i> | 1  | 2773.343  | 2773.343 | 0.773 | 0.079          | 0.688  |
| Residuals        | 9  | 32269.902 | 3585.545 |       | 0.921          |        |
| Total            | 10 | 35043.246 |          |       | 1.000          |        |

Abbreviations: PD, post-natal day; Df, degrees of freedom; SS, sum of squares; MS, mean squares; F, F-test; R<sup>2</sup>, R-squared; Pr(>F), *P*-value associated with the F-statistic

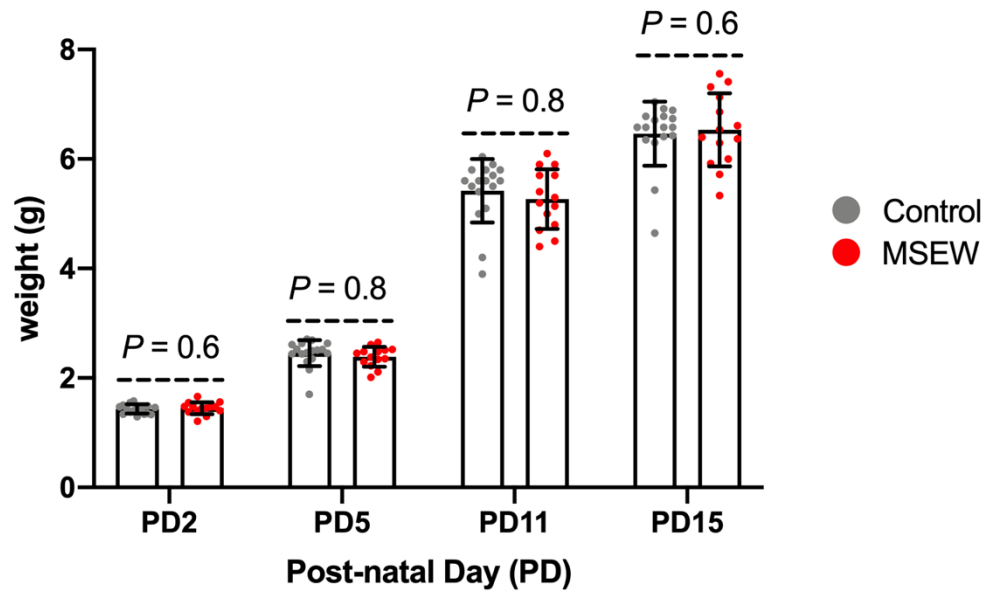
**Table S12.** Analysis of variance table for the effect of treatment on similarity metrics of pup and dam microbiota using the generalized linear mixed model (GLMM) framework. The model formula in the R language was: `lmer(similarity metric ~ treatment + (1|dam identity))`. Denominator degrees of freedom and *P*-values for the F test were approximated with the `lmerTest` package.

| PD10               |          |          |       |        |        |         |
|--------------------|----------|----------|-------|--------|--------|---------|
| No. shared ASVs    | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)  |
| <i>treatment</i>   | 93.123   | 93.123   | 1     | 23     | 0.46   | 0.505   |
| RA of shared ASVs* | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)  |
| <i>treatment</i>   | 1.023    | 1.023    | 1     | 21.576 | 0.448  | 0.510   |
| Aitchison Distance | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)  |
| <i>treatment</i>   | 1100.506 | 1100.506 | 1     | 19.925 | 12.705 | 0.002   |
| PD28               |          |          |       |        |        |         |
| No. shared ASVs    | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)  |
| <i>treatment</i>   | 2579.738 | 2579.738 | 1     | 20.262 | 27.631 | 3.7e-05 |
| RA of shared ASVs* | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)  |
| <i>treatment</i>   | 2.813    | 2.813    | 1     | 18.532 | 2.5    | 0.131   |
| Aitchison Distance | SS       | MS       | NumDf | DenDf  | F      | Pr(>F)  |
| <i>treatment</i>   | 137.425  | 137.425  | 1     | 15.048 | 4.991  | 0.041   |

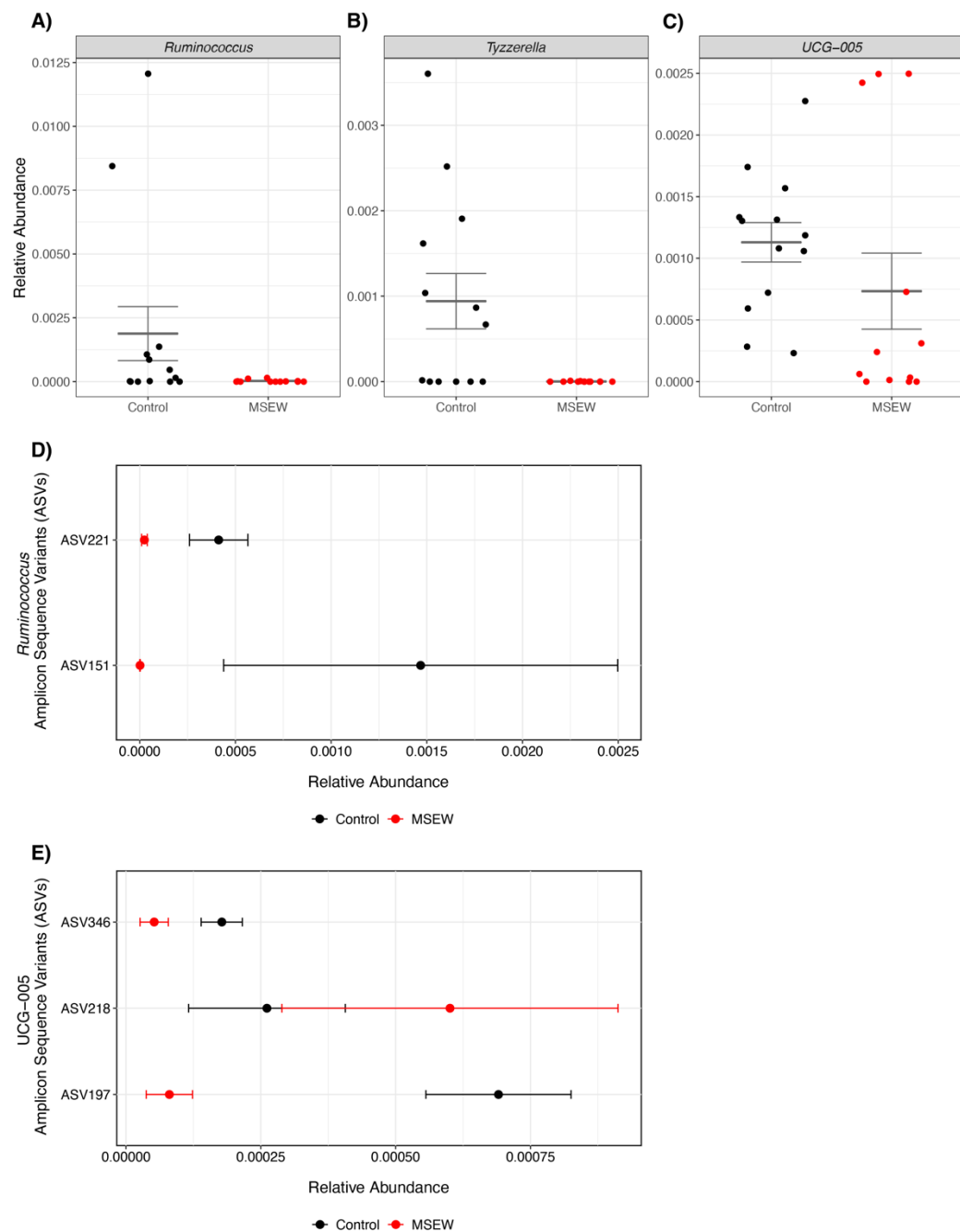
Abbreviations: PD, post-natal day; ASV, amplicon sequence variant; RA, relative abundance; SS, sum of squares; MS, mean squares; NumDf, numerator degrees of freedom; DenDf, denominator degrees of freedom; F, F-test; R<sup>2</sup>, R-squared; Pr(>F), *P*-value associated with the F-statistic

\* Proportional data was logit-transformed prior to statistical analyses.

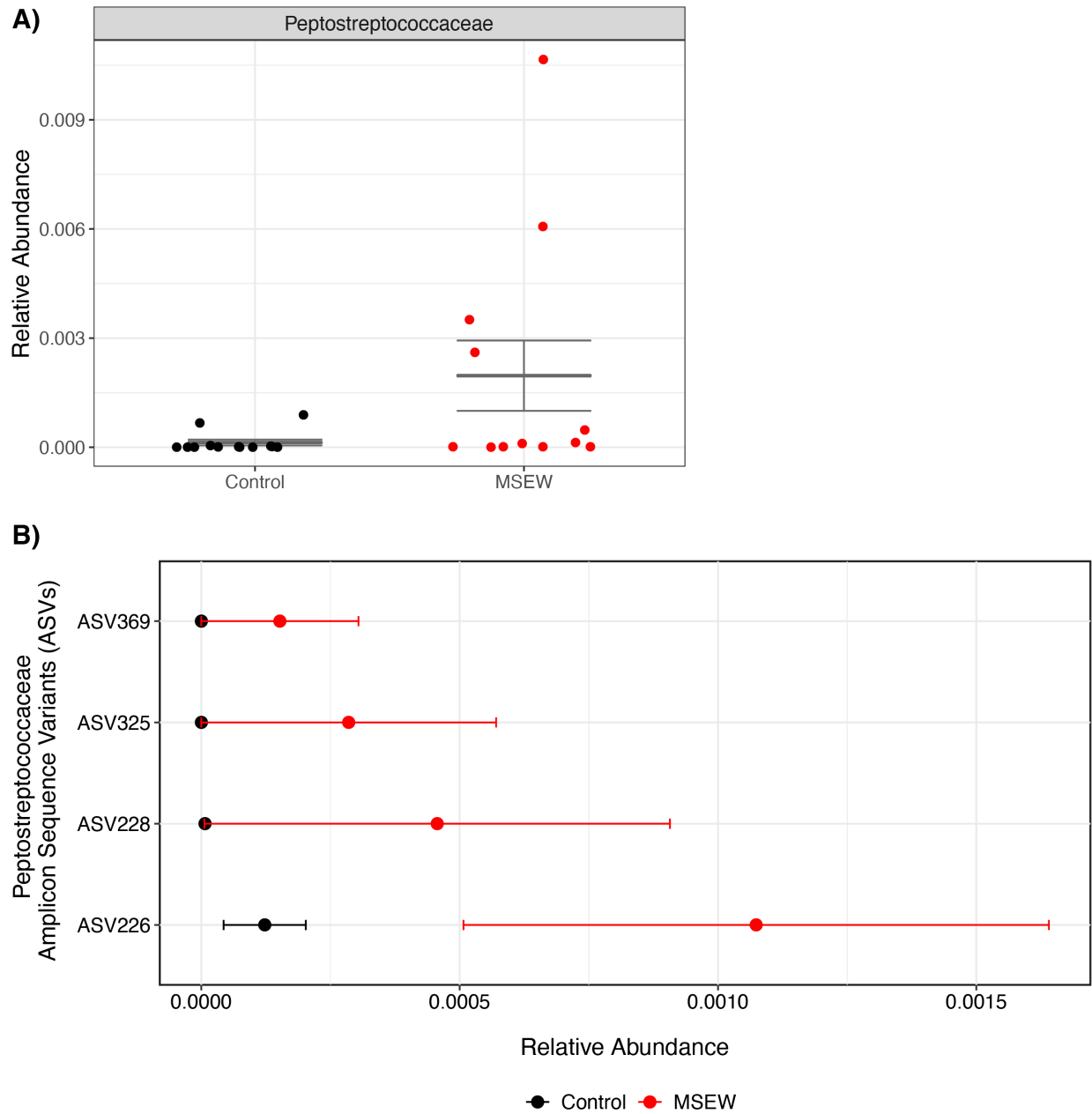
## Supplemental Figures



**Fig. S1** Body weight data (Mean  $\pm$  SEM) of normally reared control pups ( $n = 17$ ) and maternally separated with early weaning pups (MSEW,  $n = 14$ ) during post-natal (PD) time points occurring during the separation protocol.  $P$ -values are reported from generalized linear mixed models (GLMMs) conducting at each PD timepoint that adjusted for dam identity (or litter) as a random factor to assess the relationship between body weight and treatment.



**Fig. S2** Analysis of composition of microbiomes (ANCOM) at the genus level detected (A) *Ruminococcus*, (B) *Tyzzerella*, and (C) Oscillospiraceae UCG-005 as significantly enriched in PD28 normally-reared control animals compared to MSEW animals at PD28 (Mean ( $\pm$  SEM) relative abundance). Mean ( $\pm$  SEM) relative abundance of all ASVs classified as (D) *Ruminococcus* and (E) Oscillospiraceae UCG-005. The genus *Tyzzerella* comprised only one ASV in this dataset.



**Fig. S3** Analysis of composition of microbiomes (ANCOM) at the family level detected Peptostreptococcaceae as significantly enriched in PD28 MSEW animals, though no individual amplicon sequence variants (ASVs) were significantly different alone. Mean ( $\pm$  SEM) relative abundance of (A) Peptostreptococcaceae and (B) all ASVs classified as Peptostreptococcaceae.