**Colonic microbiome profiles for improved feed efficiency can be identified despite major effects of farm of origin and contemporary group in pigs**

Stafford Vigors1, John V. O’ Doherty1, Torres Sweeney2

1 *School of Agriculture & Food Science, University College Dublin, Belfield, Dublin 4, Ireland*

*2 School of Veterinary Medicine, University College Dublin, Belfield, Dublin 4, Ireland*

\* Corresponding author: torres.sweeney@ucd.ie

***animal* Journal**

*Supplementary Table S1. Composition and chemical analysis of experimental diets fed to pigs (as fed basis g/kg)*

|  |  |  |  |
| --- | --- | --- | --- |
| Ingredient | Weaner (g/kg) | Grower (g/kg) | Finisher (g/kg) |
|  Whey permeate | 125.0 |  |  |
|  Wheat | 444.2 | 400.0 | 382.6 |
|  Barley |  | 273.5 | 250.0 |
|  Soybean meal | 142.5 | 180.0 | 195.0 |
|  Maize |  |  | 150.0 |
|  Soya oil | 65.0 | 10.0 | 18.0 |
|  Whey protein isolate | 130.0 |  |  |
|  Full-fat soyabean | 80.0 | 100.0 |  |
|  Vitamins and minerals | 5.0 | 3.0 | 2.5 |
|  Limestone |  | 11.0 | 12.5 |
|  Salt |  | 3.0 | 5.0 |
|  Dicalcium phosphate  |  | 12.5 |  |
|  Monocalcium phosphate |  |  | 6.6 |
|  Lysine HCl | 4.5 | 4.0 | 2.3 |
|  DL-methionine | 1.6 | 1.5 |  |
|  L-threonine | 2.5 | 1.5 | 0.5 |
| **Analysis (g/kg, unless otherwise stated)** |
|  DM  | 892.5 | 881.1 | 884.2 |
|  CP (N x 6.25) | 224.2 | 195.0 | 193.8 |
|  Gross Energy (MJ/kg) | 18.2 | 16.5 | 16.4 |
|  Digestible Energy (MJ/kg) | 16.0 | 14.5 | 14.0 |
|  Ash | 43.7 | 50.0 | 49.8 |
|  Neutral-detergent fibre | 110.3 | 139.2 | 130.5 |
|  Lysine**†** | 16.5 | 13.1 | 11.1 |
|  Methionine and cysteine**†** | 9.90 | 8.02 | 6.60 |
|  Threonine**†** | 10.7 | 8.09 | 7.7 |
|  Tryptophan**†** | 2.5 | 2.3 | 2.0 |
|  Calcium**†** | 8.0 | 9.5 | 5.8 |
|  Phosphorous**†** | 6.0 | 6.1 | 5.8 |

Weaner diet provided (mg/kg diet): Cu 175, Fe 140, Mn 47, Zn 120, I 0.6, Se 0.3, retinol 1.8, cholecalciferol 0.025, α-tocopherol 67, phytylmenaquinone 4, cyanocobalamin 0.01, riboflavin 2, nicotinic acid 12, pantothenic acid 10, choline chloride 250, thiamine 2, pyridoxine 0.015

Grower diet provided (mg/kg diet): Cu 100, Fe 140, Mn 47, Zn 120, I 0.6, Se 0.3, retinol 1.8, cholecalciferol 0.025, α-tocopherol 67, phytylmenaquinone 4, cyanocobalamin 0.01, riboflavin 2, nicotinic acid 12, pantothenic acid 10, choline chloride 250, thiamine 2, pyridoxine 0.015

Finisher diet provided (mg/kg diet) Cu 25, Zn 100, Se 0.3, Fe 100, Mn 25, I 0.2, retinol 4.2, cholecalciferol 0.07, α-tocopherol 80

† Calculated for tabulated nutritional composition (Sauvant, 2004)

*Supplementary Table S2.* *Differential abundance analysis of all measured microbial populations from pigs divergent in residual feed intake (RFI) and the effect of selection from two different farms of birth*

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Farm |  | RFI |  | Adjusted *P-*value |
|  | A | B | SEM | High | Low | SEM | Farm1 | RFI2,3 |
| **Phylum** |   |   |   |   |   |   |   |   |
| Actinobacteria | 0.28 | 0.07 | 0.03 | 0.21 | 0.1 | 0.03 | 0.000 | 0.046 |
| Bacteroidetes | 40.2 | 65.3 | 2.51 | 54.42 | 55.62 | 2.51 | 0.01 | 0.921 |
| Cyanobacteria | 0 | 0.05 | 0.01 | 0.03 | 0.03 | 0.01 | 0.000 | 0.983 |
| Deferribacteres | 0.01 | 0.19 | 0.03 | 0.18 | 0.05 | 0.03 | 0.000 | 0.017 |
| Elusimicrobia | 0 | 0.01 | 0 | 0.01 | 0 | 0 | 0.000 | 0.315 |
| Fibrobacteres | 0.1 | 0.08 | 0.01 | 0.09 | 0.08 | 0.01 | 0.000 | 0.454 |
| Firmicutes | 56.12 | 25.74 | 2.8 | 39.02 | 37.35 | 2.8 | 0.000 | 0.885 |
| Lentisphaerae | 0 | 0.04 | 0.01 | 0.03 | 0.02 | 0.01 | 0.000 | 0.927 |
| Proteobacteria | 1.17 | 5.53 | 0.45 | 4.06 | 3.41 | 0.45 | 0.000 | 0.885 |
| Spirochaetes | 2.06 | 2.55 | 0.43 | 1.7 | 3.04 | 0.43 | 0.04 | 0.017 |
| Tenericutes | 0.04 | 0.35 | 0.04 | 0.21 | 0.23 | 0.04 | 0.000 | 0.885 |
| Verrucomicrobia | 0.01 | 0.08 | 0.01 | 0.05 | 0.05 | 0.01 | 0.000 | 0.921 |
| **Family** |  |  |  |  |  |  |  |  |
| *Alcaligenaceae* | 0.07 | 0.68 | 0.07 | 0.48 | 0.38 | 0.07 | 0.000 | 0.983 |
| *Anaeroplasmataceae* | 0.04 | 0.31 | 0.04 | 0.18 | 0.21 | 0.04 | 0.000 | 0.983 |
| *Bacillaceae* | 0.03 | 0 | 0.01 | 0.02 | 0.01 | 0.01 | 0.061 | 0.983 |
| *Bacteroidaceae* | 0.14 | 0.52 | 0.13 | 0.23 | 0.51 | 0.13 | 0.177 | 0.086 |
| *BS11* | 0.08 | 0.07 | 0.03 | 0.01 | 0.14 | 0.03 | 0.177 | 0.029 |
| *Campylobacteraceae* | 0.42 | 1.95 | 0.27 | 1.31 | 1.29 | 0.27 | 0.000 | 0.983 |
| *Christensenellaceae* | 0.47 | 0.36 | 0.13 | 0.23 | 0.61 | 0.13 | 0.449 | 0.276 |
| *Clostridiaceae* | 5.04 | 2.81 | 0.32 | 3.86 | 3.73 | 0.32 | 0.000 | 0.983 |
| *Coriobacteriaceae* | 0.3 | 0.07 | 0.03 | 0.23 | 0.11 | 0.03 | 0.000 | 0.186 |
| *Deferribacteraceae* | 0.01 | 0.22 | 0.04 | 0.2 | 0.06 | 0.04 | 0.000 | 0.086 |
| *Desulfovibrionaceae* | 0.19 | 0.13 | 0.01 | 0.16 | 0.16 | 0.01 | 0.001 | 0.983 |
| *Elusimicrobiaceae* | 0 | 0.02 | 0 | 0.01 | 0.01 | 0 | 0.000 | 0.607 |
| *Enterobacteriaceae* | 0.13 | 0.04 | 0.03 | 0.1 | 0.06 | 0.03 | 0.011 | 0.983 |
| *Erysipelotrichaceae* | 0.01 | 0.01 | 0 | 0.01 | 0.01 | 0 | 0.718 | 0.983 |
| *Fibrobacteraceae* | 0.1 | 0.1 | 0.01 | 0.11 | 0.09 | 0.01 | 0.177 | 0.983 |
| *Helicobacteraceae* | 0.02 | 0.24 | 0.03 | 0.17 | 0.14 | 0.03 | 0.969 | 0.983 |
| *Lachnospiraceae* | 19.22 | 7.54 | 1.14 | 13.79 | 11.26 | 1.14 | 0.000 | 0.983 |
| *Lactobacillaceae* | 8.11 | 1.49 | 1.13 | 3.08 | 5.59 | 1.13 | 0.000 | 0.503 |
| *Moraxellaceae* | 0 | 0.01 | 0 | 0.01 | 0 | 0 | 0.969 | 0.983 |
| *Mycoplasmataceae* | 0.02 | 0.03 | 0.01 | 0.02 | 0.02 | 0.01 | 0.018 | 0.983 |
| *p-2534-18B5* | 0.09 | 0.18 | 0.04 | 0.16 | 0.12 | 0.04 | 0.449 | 0.983 |
| *Pasteurellaceae* | 0.01 | 0.04 | 0.01 | 0.03 | 0.03 | 0.01 | 0.034 | 0.983 |
| *Peptococcaceae* | 0.04 | 0 | 0.01 | 0.02 | 0.02 | 0.01 | 0.000 | 0.983 |
| *Peptostreptococcaceae* | 0.03 | 0.01 | 0 | 0.02 | 0.02 | 0 | 0.000 | 0.983 |
| *Porphyromonadaceae* | 0.35 | 0.57 | 0.05 | 0.47 | 0.5 | 0.05 | 0.271 | 0.983 |
| *Prevotellaceae* | 31.91 | 55.71 | 2.78 | 46.12 | 44.51 | 2.78 | 0.017 | 0.983 |
| *R4-45B* | 0 | 0.04 | 0.01 | 0.03 | 0.02 | 0.01 | 0.000 | 0.983 |
| *RF16* | 0.1 | 2.85 | 0.8 | 1.14 | 2.32 | 0.8 | 0.000 | 0.983 |
| *RFP12* | 0.01 | 0.09 | 0.01 | 0.06 | 0.06 | 0.01 | 0.000 | 0.983 |
| *Ruminococcaceae* | 14.01 | 8.02 | 0.74 | 10.78 | 10.52 | 0.74 | 0.000 | 0.983 |
| *S24-7* | 2.75 | 1.22 | 0.19 | 1.55 | 2.27 | 0.19 | 0.000 | 0.186 |
| *Spirochaetaceae* | 2.22 | 2.85 | 0.49 | 1.9 | 3.42 | 0.49 | 0.695 | 0.222 |
| *Streptococcaceae* | 3.7 | 1.46 | 0.64 | 2.75 | 2.18 | 0.64 | 0.001 | 0.207 |
| *Succinivibrionaceae* | 0.21 | 1.3 | 0.14 | 0.9 | 0.78 | 0.14 | 0.000 | 0.983 |
| *Turicibacteraceae* | 0.31 | 0.48 | 0.05 | 0.46 | 0.36 | 0.05 | 0.125 | 0.983 |
| *Veillonellaceae* | 2.27 | 1.62 | 0.18 | 2.13 | 1.52 | 0.18 | 0.449 | 0.983 |
| **Genus**  |  |  |  |  |  |  |  |  |
| *[Prevotella]* | 7.22 | 6.6 | 0.48 | 7.27 | 6.25 | 0.48 | 0.025 | 0.941 |
| *Actinobacillus* | 0.02 | 0.04 | 0.01 | 0.03 | 0.03 | 0.01 | 0.001 | 0.941 |
| *Anaerobiospirillum* | 0.01 | 0.07 | 0.01 | 0.05 | 0.04 | 0.01 | 0.000 | 0.941 |
| *Anaeroplasma* | 0.04 | 0.24 | 0.04 | 0.12 | 0.19 | 0.04 | 0.000 | 0.83 |
| *Anaerostipes* | 0.03 | 0.01 | 0 | 0.02 | 0.01 | 0 | 0.000 | 0.891 |
| *Anaerovibrio* | 1.56 | 1.26 | 0.2 | 1.34 | 1.29 | 0.2 | 0.154 | 0.941 |
| *Bacteroides* | 0.21 | 0.72 | 0.21 | 0.29 | 0.75 | 0.21 | 0.014 | 0.064 |
| *Blautia* | 0.31 | 0.19 | 0.02 | 0.28 | 0.21 | 0.02 | 0.331 | 0.649 |
| *Butyrivibrio* | 0.09 | 0.23 | 0.12 | 0.29 | 0.05 | 0.12 | 0.027 | 0.941 |
| *Campylobacter* | 0.58 | 2.41 | 0.34 | 1.63 | 1.64 | 0.34 | 0.000 | 0.941 |
| *CF231* | 2.32 | 1.38 | 0.22 | 1.39 | 2.2 | 0.22 | 0.688 | 0.079 |
| *Clostridium* | 1.22 | 0.41 | 0.12 | 0.8 | 0.73 | 0.12 | 0.000 | 0.97 |
| *Collinsella* | 0.17 | 0.04 | 0.02 | 0.15 | 0.05 | 0.02 | 0.000 | 0.038 |
| *Coprococcus* | 4.32 | 0.94 | 0.36 | 2.68 | 2.09 | 0.36 | 0.000 | 0.912 |
| *Desulfovibrio* | 0.25 | 0.1 | 0.02 | 0.17 | 0.17 | 0.02 | 0.000 | 0.941 |
| *Dialister* | 0.75 | 0.43 | 0.16 | 0.87 | 0.26 | 0.16 | 0.711 | 0.432 |
| *Dorea* | 0.32 | 0.15 | 0.02 | 0.24 | 0.21 | 0.02 | 0.001 | 0.941 |
| *Faecalibacterium* | 2.86 | 0.71 | 0.28 | 2.11 | 1.07 | 0.28 | 0.000 | 0.334 |
| *Fibrobacter* | 0.15 | 0.12 | 0.01 | 0.14 | 0.13 | 0.01 | 0.716 | 0.941 |
| *Helicobacter* | 0.03 | 0.23 | 0.03 | 0.16 | 0.13 | 0.03 | 0.000 | 0.941 |
| *Lachnospira* | 1.24 | 0.94 | 0.09 | 1.11 | 1 | 0.09 | 0.366 | 0.941 |
| *Lactobacillus* | 11.55 | 1.8 | 1.63 | 4.13 | 7.9 | 1.63 | 0.000 | 0.298 |
| *Megasphaera* | 0.16 | 0.01 | 0.02 | 0.11 | 0.04 | 0.02 | 0.000 | 0.432 |
| *Mitsuokella* | 0.48 | 0.17 | 0.06 | 0.34 | 0.26 | 0.06 | 0.199 | 0.941 |
| *Mogibacterium* | 0 | 0.02 | 0.01 | 0 | 0.02 | 0.01 | 0.002 | 0.432 |
| *Mucispirillum* | 0.01 | 0.26 | 0.04 | 0.24 | 0.07 | 0.04 | 0.000 | 0.244 |
| *Mycoplasma* | 0.02 | 0.04 | 0.01 | 0.03 | 0.02 | 0.01 | 0.008 | 0.941 |
| *Oscillospira* | 2.12 | 0.94 | 0.17 | 1.3 | 1.63 | 0.17 | 0.009 | 0.432 |
| *p-75-a5* | 0.01 | 0.01 | 0 | 0.01 | 0.01 | 0 | 0.149 | 0.95 |
| *Paludibacter* | 0 | 0.11 | 0.03 | 0.08 | 0.05 | 0.03 | 0.000 | 0.912 |
| *Parabacteroides* | 0.49 | 0.61 | 0.06 | 0.51 | 0.63 | 0.06 | 0.043 | 0.891 |
| *Phascolarctobacterium* | 0.07 | 0.04 | 0.01 | 0.04 | 0.06 | 0.01 | 0.668 | 0.298 |
| *Prevotella* | 42.4 | 67.24 | 2.93 | 57.28 | 55.67 | 2.93 | 0.000 | 0.941 |
| *Psychrobacter* | 0 | 0.01 | 0.01 | 0.01 | 0 | 0.01 | 0.904 | 0.941 |
| *rc4-4* | 0.07 | 0.01 | 0.01 | 0.03 | 0.04 | 0.01 | 0.000 | 0.733 |
| *Roseburia* | 5.47 | 2.02 | 0.46 | 4.16 | 2.81 | 0.46 | 0.002 | 0.559 |
| *Ruminococcus* | 3.46 | 0.9 | 0.31 | 1.76 | 2.32 | 0.31 | 0.000 | 0.432 |
| *Selenomonas* | 0.08 | 0.07 | 0.03 | 0.08 | 0.08 | 0.03 | 0.128 | 0.891 |
| *Shuttleworthia* | 0.18 | 0.13 | 0.03 | 0.18 | 0.13 | 0.03 | 0.882 | 0.941 |
| *Slackia* | 0.03 | 0 | 0.01 | 0.03 | 0.01 | 0.01 | 0.000 | 0.149 |
| *SMB53* | 0.05 | 0.05 | 0 | 0.05 | 0.05 | 0 | 0.009 | 0.941 |
| *Streptococcus* | 5.11 | 1.82 | 0.89 | 3.81 | 2.76 | 0.89 | 0.029 | 0.941 |
| *Succinivibrio* | 0.27 | 1.25 | 0.13 | 0.85 | 0.81 | 0.13 | 0.000 | 0.941 |
| *Sutterella* | 0.09 | 0.82 | 0.08 | 0.59 | 0.46 | 0.08 | 0.000 | 0.941 |
| *Treponema* | 3.36 | 3.67 | 0.73 | 2.42 | 4.89 | 0.73 | 0.331 | 0.298 |
| *Turicibacter* | 0.46 | 0.57 | 0.07 | 0.6 | 0.46 | 0.07 | 0.002 | 0.941 |
| *YRC22* | 0.36 | 0.17 | 0.05 | 0.21 | 0.31 | 0.05 | 0.237 | 0.559 |
| **Species** |  |  |  |  |  |  |  |  |
| *Bacteroides plebeius* | 0 | 0.03 | 0.01 | 0.01 | 0.03 | 0.01 | 0.000 | 0.222 |
| *Clostridium neonatale* | 0.05 | 0.05 | 0.01 | 0.04 | 0.06 | 0.01 | 0.279 | 0.222 |
| *Collinsella aerofaciens* | 0.6 | 0.1 | 0.06 | 0.38 | 0.22 | 0.06 | 0.022 | 0.077 |
| *Coprococcus eutactus* | 1.46 | 0.16 | 0.13 | 0.62 | 0.76 | 0.13 | 0.000 | 0.669 |
| *Dorea formicigenerans* | 0.14 | 0.04 | 0.02 | 0.1 | 0.06 | 0.02 | 0.895 | 0.222 |
| *Faecalibacterium prausnitzii* | 9.3 | 1.74 | 0.82 | 5.8 | 3.84 | 0.82 | 0.02 | 0.622 |
| *Lactobacillus agilis* | 0.46 | 0.02 | 0.07 | 0.17 | 0.23 | 0.07 | 0.001 | 0.843 |
| *Lactobacillus mucosae* | 0.39 | 0.07 | 0.1 | 0.1 | 0.31 | 0.1 | 0.692 | 0.222 |
| *Lactobacillus ruminis* | 0.23 | 0.01 | 0.04 | 0.08 | 0.11 | 0.04 | 0.000 | 0.222 |
| *Mucispirillum schaedleri* | 0.05 | 0.7 | 0.11 | 0.56 | 0.3 | 0.11 | 0.000 | 0.222 |
| *Prevotella copri* | 58.64 | 77.09 | 2.9 | 70.75 | 68.22 | 2.9 | 0.000 | 0.669 |
| *Prevotella stercorea* | 8.52 | 12.75 | 0.99 | 9.65 | 12.45 | 0.99 | 0.000 | 0.222 |
| *Ruminococcus flavefaciens* | 4.9 | 0.43 | 0.85 | 0.9 | 3.7 | 0.85 | 0.000 | 0.022 |
| *Streptococcus luteciae* | 15.27 | 6.83 | 2.68 | 10.85 | 9.7 | 2.68 | 0.362 | 0.843 |

1 Significance refers to the comparison of farm A vs B

2 Significance refers to the comparison of low vs high residual feed intake (RFI). The analysis was based on pigs selected from two farms with 8 HRFI and LRFI pigs from farm A and 12 HRFI and LRFI pigs from farm B

3 No interaction was identified between farm of birth and RFI so these data are not presented.

*Supplementary Table S3. Analysis of the influence of residual feed intake and selection from two different farms of birth on colonic volatile fatty acid (VFA) profiles in pigs*

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
|  | Farm |   | RFI |   | Adjusted *P*-value |
|   | A | B | SEM | High | Low | SEM | Farm1 | RFI2,3 |
| Total (mmol/kg DM) | 92.30 | 220.97 | 10.49 | 177.16 | 159.66 | 10.49 | 0.001 | 0.156 |
| Acetic | 54.82 | 160.80 | 8.65 | 123.52 | 111.44 | 8.65 | 0.001 | 0.381 |
| Propionic | 15.90 | 30.98 | 1.35 | 26.01 | 23.64 | 1.35 | 0.001 | 0.281 |
| Butyric | 16.32 | 18.51 | 0.54 | 18.67 | 16.61 | 0.54 | 0.042 | 0.673 |
| Valeric | 2.70 | 4.73 | 0.25 | 4.07 | 3.73 | 0.25 | 0.001 | 0.585 |
| Iso-Butyric | 1.49 | 0.80 | 0.12 | 0.97 | 1.15 | 0.12 | 0.001 | 0.344 |
| iso-Valeric | 1.35 | 5.15 | 0.38 | 4.20 | 3.16 | 0.38 | 0.001 | 0.356 |
| Ratio to total VFA |  |  |  |  |  |  |  |
| Acetic | 0.60 | 0.73 | 0.01 | 0.68 | 0.67 | 0.01 | 0.001 | 0.934 |
| Propionic | 0.17 | 0.14 | 0.00 | 0.15 | 0.16 | 0.00 | 0.001 | 0.965 |
| Butyric | 0.18 | 0.08 | 0.01 | 0.12 | 0.12 | 0.01 | 0.001 | 0.804 |
| Valeric | 0.03 | 0.02 | 0.00 | 0.02 | 0.02 | 0.00 | 0.014 | 0.938 |

1 Significance refers to the comparison of farm A vs B

2 Significance refers to the comparison of low vs high residual feed intake (RFI). The analysis was based on pigs selected from two farms with 8 HRFI and LRFI pigs from farm A and 12 HRFI and LRFI pigs from farm B

3 No interaction was identified between farm of birth and RFI so these data are not presented.