Supplementary Material

**Table S1 Composition of basal diets and nutrient levels (as-fed basis)**

|  |  |  |
| --- | --- | --- |
| Items | Early nursery feed (28-48 days of age) | Late nursery feed (49-69 days of age) |
| Ingredients |  |
| Corn | 22.00 | 69.50 |
| Broken rice | 25.00 | - |
| Wheat flour | 12.00 | - |
| Glucose | 3.00 | - |
| Soybean meal (46% CP) | 10.50 | - |
| Soybean meal (43% CP) | - | 16.00 |
| Puffed soybean | 10.00 | - |
| Fermented soybean | 2.50 | 4.00 |
| Soybean protein concentrate | - | 2.00 |
| Fish meal | 3.00 | 1.00 |
| Low-protein whey power | 5.00 | - |
| Egg power | 0.50 | - |
| Wheat bran | - | 2.00 |
| Soybean oil | 1.00 | 1.50 |
| Citric acid | 1.50 | - |
| Early nursery Premix 1) | 4.00 | - |
| Late nursery Premix 2) | - | 4.00 |
| Total | 100.00 | 100.00 |
| Nutrient level 3) | - | - |
| Energy (MJ/Kg) | 14.24 | 13.83 |
| Crude protein | 24.50 | 20.65 |
| Crude fat | 3.10 | 2.33 |
| Calcium | 0.81 | 0.97 |
| Total phosphorus | 0.53 | 0.55 |
| Lysine | 1.36 | 1.18 |
| Methionine | 0.50 | 0.39 |
| Threonine | 0.87 | 0.72 |

1),2) The premix compositions were in accordance with NRC (2012) recommended nutrient requirements for growing and fattening pigs. 3) Energy is a calculated value, while the others are measured values.

**Table S2. Primers for real-time PCR analysis**

|  |  |  |
| --- | --- | --- |
| Gene name | Sequence (5'-3') | Product size (bp) |
| *E-cadherin* | F:GAAGGAGGTGGAGAAGAGGAC | R:AGAGTCATAAGGTGGGGCAGT | 216  |
| *IL-1β* | F: GCTAACTACGGTGACAACAA | R: TCTTCATCGGCTTCTCCACT | 196  |
| *IL-2* | F: TGCACTAACCCTTGCACTCA | R: CAACTGTAAATCCAGCAGCAA | 100  |
| *IL-6* | F: TCCAGCATCATTGCATCATC | R: GGCTCCACTCACTCCACAAG | 105  |
| *IL-10* | F:GGGCTATTTGTCCTGACTGC | R:GGGCTCCCTAGTTTCTCTTCC | 105  |
| *IFN-α* | F:CCTGGACCACAGAAGGGA | R:TCTCATGCACCAGAGCCA | 92  |
| *Occludin* | F:ATGCCTCCTCCCCTTTCG | R:CGCCCGTCGTGTAGTCTGTC | 295  |
| *TLR-4* | F:CAGATAAGCGAGGCCGTCATT | R:TTGCAGCCCACAAAAAGCA | 113  |
| *TNFα* | F: ACAGGCCAGCTCCCTCTTAT | R: CCTCGCCCTCCTGAATAAAT | 102  |
| *ZO-1* | F:TACCCTGCGGCTGGAAGA | R:GGACGGGACCTGCTCATAACT | 154  |
| *β-actin* | F: CTGCGGCATCCACGAAACT | R: AGGGCCGTGATCTCCTTCTG | 147  |

**Table S3 Effect of dietary BLS mix supplementation on gut metabolites in weaned piglets on day 7** **of trial**

|  |  |  |  |
| --- | --- | --- | --- |
| Items | Control | Antibiotics | Probiotics |
| Short-chain fatty acids (mg/g) |
| Acetate | 3.12±0.17 | 2.86±0.17 | 2.85±0.16 |
| Butyrate | 0.80±0.06 | 0.86±0.08 | 0.92±0.10 |
| Isobutyrate | 0.07±0.01 | 0.07±0.01 | 0.08±0.01 |
| Isovalerate | 0.11±0.02 | 0.13±0.01 | 0.14±0.01 |
| Propionate | 1.12±0.03 | 1.28±0.13 | 1.33±0.14 |
| Valerat | 0.18±0.05 | 0.10±0.02 | 0.12±0.02 |
| Bioamines (μg/g) |
| Indole  | 8.21±1.29 | 6.29±1.68 | 5.95±0.74 |
| Phenylethylamine  | 0.28±0.10 | 0.36±0.09 | 0.08±0.04 |
| Putrescine | 1.82±1.01 | 0.75±0.22 | 2.37±1.41 |
| Skatole  | 3.99±0.98 | 5.08±0.61 | 3.24±0.28 |
| Spermidine  | 2.98±0.38 | 3.22±0.46 | 2.53±0.90 |
| Spermine | 6.16±2.03 | 2.15±1.62 | 4.25±1.93 |
| Tryptamine  | 0.09±0.06 | 0.03±0.01 | 0.10±0.05 |
| Tyramine | 1.15±0.53 | 0.87±0.25 | 1.34±0.61 |

**Table S4 Effect of dietary BLS mix supplementation on intestinal health-related genes in weaned piglets**

|  |  |  |
| --- | --- | --- |
| Items | Jejunum | Ileum |
| Control | Antibiotics | Probiotics | Control | Antibiotics | Probiotics |
| Day 7 |  |
| E-cadherin | 1.00±0.10 | 0.95±0.23 | 1.24±0.18 | 1.00±0.39 | 0.51±0.05 | 1.07±0.07 |
| IL-2 | 1.00±0.14a | 0.56±0.11b | 1.06±0.11a | 1.00±0.15b | 0.82±0.18b | 1.31±0.15a |
| IL-6 | 1.00±0.07 | 1.16±0.10 | 1.16±0.15 | 1.00±0.14ab | 0.80±0.06b | 1.16±0.07a |
| IL-10 | 1.00±0.11 | 1.19±0.19 | 1.37±0.16 | 1.00±0.18 | 0.81±0.07 | 1.31±0.27 |
| IL-1β | 1.00±0.10 | 1.48±0.35 | 2.45±0.53 | 1.00±0.16ab | 0.63±0.10b | 2.21±0.90a |
| IFN-α | 1.00±0.13 | 1.29±0.26 | 1.07±0.06 | 1.00±0.46 | 0.89±0.08 | 1.72±0.34 |
| Occludin | 1.00±0.12 | 1.51±0.13 | 1.65±0.22 | 1.00±0.11 | 1.30±0.26 | 1.61±0.30 |
| TLR-4 | 1.00±0.03 | 1.26±0.14 | 0.91±0.04 | 1.00±0.07b | 0.91±0.06b | 2.28±0.47a |
| TNF-α | 1.00±0.11 | 1.20±0.20 | 1.04±0.31 | 1.00±0.10ab | 1.58±0.39a | 0.80±0.11b |
| ZO-1 | 1.00±0.09 | 1.16±0.12 | 1.17±0.10 | 1.00±0.13 | 1.03±0.04 | 1.76±0.47 |
| Day 21 |  |
| E-cadherin | 1.00±0.17 | 0.65±0.14 | 1.25±0.20 | 1.00±0.06 | 0.92±0.09 | 1.30±0.20 |
| IL-2 | 1.00±0.09 | 1.46±0.31 | 1.00±0.09 | 1.00±0.24 | 1.30±0.29 | 1.15±0.21 |
| IL-6 | 1.00±0.25 | 0.70±0.13 | 1.06±0.06 | 1.00±0.07 | 1.20±0.16 | 1.08±0.10 |
| IL-10 | 1.00±0.29 | 0.96±0.33 | 1.03±0.12 | 1.00±0.08 | 1.05±0.14 | 1.91±0.25 |
| IL-1β | 1.00±0.34 | 0.91±0.22 | 1.48±0.33 | 1.00±0.07 | 0.77±0.13 | 1.26±0.37 |
| IFN-α | 1.00±0.13 | 0.90±0.21 | 1.18±0.12 | 1.00±0.14 | 1.09±0.27 | 1.05±0.14 |
| Occludin | 1.00±0.11 | 1.04±0.18 | 1.02±0.09 | 1.00±0.04 | 0.85±0.16 | 1.15±0.23 |
| TLR-4 | 1.00±0.52 | 1.12±0.52 | 0.84±0.06 | 1.00±0.22ab | 0.80±0.08b | 1.69±0.42a |
| TNF-α | 1.00±0.09 | 1.38±0.27 | 2.47±0.63 | 1.00±0.17b | 1.92±0.18a | 2.62±0.41a |
| ZO-1 | 1.00±0.11 | 0.94±0.12 | 1.09±0.14 | 1.00±0.05 | 1.05±0.14 | 1.28±0.18 |
| Day 42 |  |
| E-cadherin | 1.00±0.16 | 0.92±0.02 | 0.91±0.02 | 1.00±0.06 | 0.99±0.05 | 1.61±0.37 |
| IL-2 | 1.00±0.26 | 0.84±0.12 | 0.73±0.10 | 1.00±0.17 | 0.96±0.21 | 1.60±0.50 |
| IL-6 | 1.00±0.11 | 1.39±0.26 | 0.97±0.07 | 1.00±0.13 | 1.24±0.10 | 1.26±0.09 |
| IL-10 | 1.00±0.14 | 1.32±0.19 | 1.32±0.11 | 1.00±0.13 | 1.01±0.09 | 1.22±0.15 |
| IL-1β | 1.00±0.19 | 1.26±0.26 | 1.28±0.30 | 1.00±0.15 | 1.01±0.20 | 1.26±0.13 |
| IFN-α | 1.00±0.19 | 0.88±0.11 | 0.98±0.11 | 1.00±0.09 | 0.91±0.21 | 1.07±0.20 |
| Occludin | 1.00±0.43 | 1.69±1.09 | 0.57±0.05 | 1.00±0.11 | 1.16±0.17 | 1.06±0.13 |
| TLR-4 | 1.00±0.13 | 1.15±0.09 | 1.50±0.64 | 1.00±0.15b | 1.05±0.12ab | 1.72±0.42a |
| TNF-α | 1.00±0.28 | 0.88±0.18 | 1.35±0.40 | 1.00±0.13 | 1.43±0.20 | 2.88±1.30 |
| ZO-1 | 1.00±0.24 | 0.84±0.07 | 0.72±0.07 | 1.00±0.07 | 1.10±0.12 | 1.47±0.12 |