

1    **Dietary phytase and lactic acid treated cereals caused greater taxonomic than functional**  
2    **adaptations in the cecal metagenome of growing pigs**

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15   Running title: Phosphorus and cecal metagenome

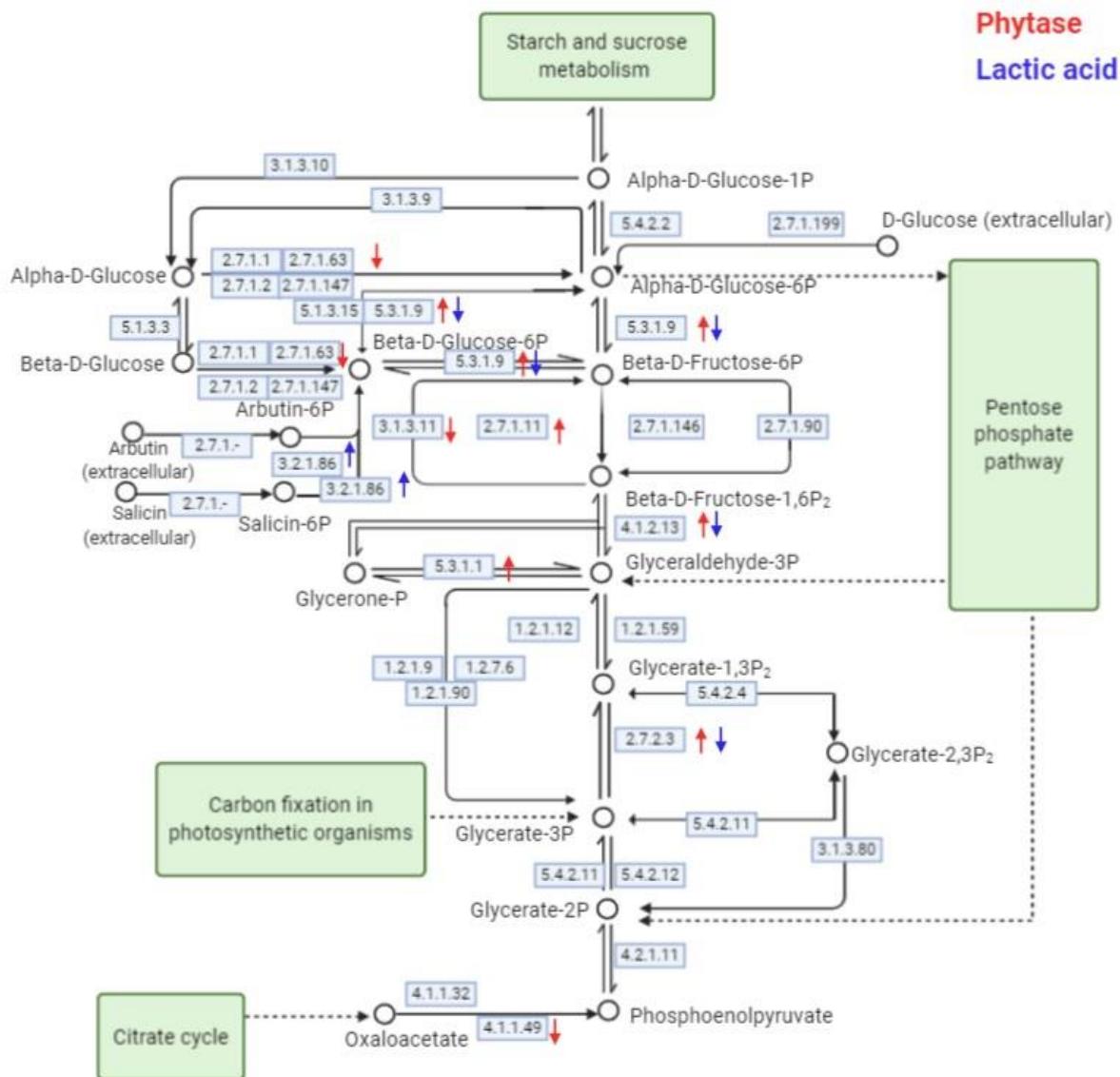
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18   KEYWORDS: cecum, metagenome, lactic acid treatment of cereal grains, pig, phytase

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## Glycolysis/Gluconeogenesis



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21 **FIG S1** Map of genes corresponding to the key enzymes related to KEGG orthology (KO)  
22 functions within ‘Glycolysis / Gluconeogenesis’ in the metagenome of pigs fed diets with or  
23 without phytase and lactic acid treatment of cereals. Arrows illustrate significant differences  
24 and tendencies for differences in gene abundances due to phytase supplementation (red) and  
25 lactic acid treatment of dietary cereals (blue). Detailed information on significant differences  
26 and tendencies for differences can be found in Table S6.

27 **TABLE S1** Ingredient and chemical composition of experimental diets

Treatment of cereal grains	No phytase		Phytase	
	Con	LA	Con	LA
Ingredient (%)				
Wheat	36.2	36.2	36.2	36.2
Corn	36.0	36.0	36.0	36.0
Soybean HP dehulled	22.0	22.0	22.0	22.0
Sunflower oil	2.0	2.0	2.0	2.0
Vitamin-mineral-premix <sup>a</sup>	2.3	2.3	2.3	2.3
Limestone	1.1	1.1	1.1	1.1
Monocalcium phosphate	0.4	0.4	0.4	0.4
Phytase (FTU/kg) <sup>b</sup>	0	0	500	500
Analyzed chemical composition (g/kg dry matter)				
Dry matter	900	942	900	942
Crude ash	54	49	54	49
Crude protein	213	202	216	205
Neutral detergent fiber	131	122	129	119
Acid detergent fiber	50	51	52	51
Hemicellulose <sup>c</sup>	81	71	77	68
Resistant starch	8.2	7.7	9.2	8.3
Non-resistant starch	511	492	500	485
Phosphorus	5.5	4.9	5.5	4.9
Calcium	6.6	6.2	6.6	6.2

28 Con, control diet; LA, lactic acid. <sup>a</sup>The vitamin-mineral-premix provided per kilogram of  
29 experimental diet (Garant GmbH, Pöchlarn, Austria): vitamin A, 17.250 IE; vitamin D3,  
30 2.299 IE; vitamin E, 160.999 mg; vitamin K3, 4.600 mg; vitamin B1, 2.299 mg; vitamin B2,  
31 6.900 mg; vitamin B6, 3.450 mg; vitamin B12, 0.034 mg; vitamin B3, 34.500 mg;  
32 Pantothenic acid, 17.150 mg; Folic acid, 1.150 mg; Biotin, 0.172 mg; Choline Chloride,  
33 579.600 mg; Fe, 103.500 mg; Cu, 17.250 mg; Zn, 103.500 mg; Mn, 45.999 mg; J, 1.725 mg;  
34 Se, 0.517 mg.

35 <sup>b</sup>FTU; phytase units.

36 <sup>c</sup>calculated as the difference between neutral detergent fiber and acid detergent fiber.

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38 **TABLE S2** Permutational multivariate analysis of variance (PERMANOVA) results for cecal  
39 bacterial communities of pigs (amplicon sequence variants at relative abundance > 0.01% of  
40 all reads) fed diets with or without phytase and lactic acid treatment of cereals

Source of Variation	df	SS	R <sup>2</sup>	F	P-value <sup>41</sup> <sub>42</sub>
Diet	3	0.14	0.152	1.677	0.041 <sup>43</sup>
Phytase	1	0.06	0.072	2.375	0.028 <sup>44</sup>
Lactic acid	1	0.02	0.025	0.837	0.518 <sup>45</sup>
Phytase : Lactic acid	1	0.05	0.055	1.817	0.100 <sup>46</sup>
Residual	28	0.75	0.848		<sup>47</sup>
Total	31	0.89	1.000		<sup>48</sup>

50 The analysis based on pairwise distance of a multivariate data set and values were obtained  
51 using type III sums of squares with 999 permutations of residuals. Significant difference was  
52 declared at  $P \leq 0.05$ . df, degrees of freedom; SS, sum of squares.

53 **TABLE S3** Bacterial genera (relative abundance > 0.1% of all reads) in cecal digesta of pigs fed diets with or without phytase and lactic acid  
 54 treatment of cereals

Treatment of cereal grains	No phytase		Phytase		SEM	P-value		
	Con	LA	Con	LA		Phytase	LA	Phytase×LA
<i>Prevotella</i>	32.40	35.23	27.42	28.19	2.035	0.007	0.385	0.616
<i>Bacteroides</i>	13.41	11.31	12.74	12.10	0.419	0.898	0.003	0.095
<i>Clostridium</i>	7.83 ab	6.19 b	8.25 ab	9.82 a	0.710	0.009	0.968	0.033
<i>Lactobacillus</i>	6.42	8.56	8.84	8.00	1.272	0.471	0.613	0.254
<i>Eubacterium</i>	4.49	4.55	5.57	5.14	0.438	0.068	0.680	0.575
<i>Ruminococcus</i>	2.53	2.12	2.65	2.91	0.226	0.055	0.747	0.145
<i>Faecalibacterium</i>	1.42	1.53	1.89	1.84	0.227	0.099	0.894	0.752
<i>Mitsuokella</i>	1.44	2.33	1.13	1.41	0.180	0.002	0.003	0.103
<i>Roseburia</i>	1.24	1.17	1.65	1.53	0.148	0.015	0.519	0.871
<i>Selenomonas</i>	1.14	1.41	1.34	1.18	0.135	0.904	0.687	0.124
<i>Parabacteroides</i>	1.36	1.02	1.36	1.29	0.075	0.083	0.011	0.088
<i>Subdoligranulum</i>	0.95	0.93	1.25	1.15	0.110	0.027	0.588	0.678
<i>Butyrivibrio</i>	0.90	0.75	1.20	1.03	0.138	0.051	0.257	0.931
<i>Bacillus</i>	0.763	0.728	0.856	0.917	0.041	0.002	0.756	0.260
<i>Acidaminococcus</i>	0.772	0.922	0.707	0.714	0.044	0.005	0.086	0.119
<i>Streptococcus</i>	0.701	0.456	0.802	0.534	0.108	0.413	0.025	0.916
<i>Megasphaera</i>	0.464	0.811	0.408	0.461	0.108	0.071	0.075	0.184
<i>Campylobacter</i>	0.626	0.777	0.510	0.207	0.155	0.037	0.631	0.156
<i>Coprococcus</i>	0.510	0.426	0.517	0.510	0.048	0.351	0.356	0.430
<i>Veillonella</i>	0.447 b	0.619 a	0.435 b	0.458 b	0.030	0.007	0.003	0.019
<i>Dorea</i>	0.466	0.435	0.504	0.506	0.030	0.077	0.635	0.586
<i>Porphyromonas</i>	0.500	0.364	0.493	0.439	0.024	0.174	0.001	0.100

<i>Bifidobacterium</i>	0.402	0.439	0.464	0.354	0.074	0.878	0.633	0.330
<i>Blautia</i>	0.392	0.300	0.493	0.443	0.052	0.028	0.183	0.694
<i>Dialister</i>	0.330	0.696	0.200	0.401	0.062	0.002	0.000	0.194
Unclassified	0.374	0.340	0.416	0.467	0.029	0.008	0.780	0.158
<i>Erysipelotrichaceae</i>								
Unclassified	0.384	0.294	0.457	0.455	0.043	0.012	0.299	0.321
<i>Lachnospiraceae</i>								
<i>Desulfitobacterium</i>	0.345	0.324	0.377	0.377	0.019	0.033	0.594	0.576
<i>Alkaliphilus</i>	0.329	0.289	0.358	0.381	0.021	0.008	0.687	0.144
Unclassified	0.357	0.219	0.371	0.362	0.033	0.025	0.037	0.064
<i>Ruminococcaceae</i>								
<i>Ethanoligenens</i>	0.313	0.259	0.341	0.340	0.025	0.035	0.267	0.287
<i>Holdemania</i>	0.260	0.242	0.307	0.341	0.019	0.001	0.673	0.191
<i>Paludibacter</i>	0.288	0.225	0.285	0.262	0.013	0.208	0.003	0.135
<i>Desulfotomaculum</i>	0.251	0.260	0.276	0.267	0.013	0.212	0.969	0.488
Unclassified <i>Clostridiales</i>	0.260	0.204	0.288	0.285	0.020	0.011	0.149	0.190
<i>Fusobacterium</i>	0.240	0.215	0.258	0.260	0.012	0.012	0.341	0.264
<i>Treponema</i>	0.230	0.097	0.255	0.381	0.091	0.102	0.970	0.168
<i>Caldicellulosiruptor</i>	0.223	0.198	0.256	0.256	0.014	0.004	0.398	0.405
<i>Paenibacillus</i>	0.209	0.192	0.235	0.243	0.014	0.012	0.733	0.383
<i>Enterococcus</i>	0.203	0.191	0.228	0.242	0.012	0.003	0.961	0.293
<i>Geobacillus</i>	0.184	0.183	0.203	0.220	0.009	0.005	0.396	0.330
<i>Fibrobacter</i>	0.213	0.173	0.208	0.195	0.011	0.435	0.025	0.254
<i>Flavobacterium</i>	0.208	0.178	0.197	0.187	0.007	0.905	0.006	0.132
<i>Desulfovibrio</i>	0.205	0.182	0.194	0.183	0.016	0.756	0.304	0.717
<i>Oribacterium</i>	0.164	0.184	0.174	0.206	0.014	0.278	0.077	0.666
<i>Slackia</i>	0.173	0.156	0.185	0.188	0.010	0.043	0.521	0.341

<i>Geobacter</i>	0.168	0.164	0.179	0.176	0.009	0.203	0.656	0.949
<i>Caldanaerobacter</i>	0.159	0.153	0.181	0.185	0.009	0.004	0.954	0.581
<i>Thermoanaerobacter</i>	0.154	0.145	0.174	0.180	0.008	0.003	0.878	0.372
<i>Catenibacterium</i>	0.104	0.150	0.101	0.224	0.033	0.287	0.017	0.254
<i>Anaerotruncus</i>	0.156	0.101	0.153	0.154	0.015	0.100	0.086	0.074
<i>Atopobium</i>	0.127	0.148	0.118	0.156	0.014	0.997	0.043	0.536
<i>Brachyspira</i>	0.134	0.124	0.139	0.145	0.008	0.094	0.796	0.311
<i>Eggerthella</i>	0.128	0.120	0.136	0.143	0.007	0.039	0.955	0.326
<i>Abiotrophia</i>	0.124	0.102	0.145	0.139	0.012	0.029	0.265	0.518
<i>Alistipes</i>	0.144	0.101	0.142	0.120	0.008	0.317	0.001	0.228
<i>Heliobacterium</i>	0.120	0.129	0.127	0.123	0.006	0.893	0.711	0.341
<i>Helicobacter</i>	0.148	0.166	0.114	0.071	0.023	0.011	0.601	0.201
<i>Shewanella</i>	0.148	0.123	0.116	0.110	0.009	0.020	0.096	0.305
<i>Pedobacter</i>	0.131	0.105	0.129	0.117	0.005	0.325	0.001	0.191
<i>Listeria</i>	0.110	0.109	0.124	0.132	0.005	0.001	0.524	0.403
<i>Anaerostipes</i>	0.119	0.089	0.130	0.131	0.014	0.070	0.317	0.278
<i>Staphylococcus</i>	0.105	0.103	0.122	0.132	0.006	0.001	0.517	0.311
<i>Carboxydothermus</i>	0.107	0.114	0.117	0.119	0.005	0.150	0.349	0.607
<i>Pelotomaculum</i>	0.107	0.117	0.114	0.114	0.005	0.660	0.311	0.306
<i>Thermosinus</i>	0.108	0.103	0.129	0.106	0.010	0.250	0.178	0.391
<i>Collinsella</i>	0.098	0.135	0.085	0.125	0.012	0.356	0.004	0.893
<i>Vibrio</i>	0.132	0.107	0.097	0.089	0.010	0.016	0.130	0.399
<i>Syntrophomonas</i>	0.098	0.094	0.112	0.108	0.005	0.014	0.473	0.990
<i>Capnocytophaga</i>	0.109 a	0.088 b	0.106 a	0.104 a	0.004	0.150	0.011	0.038
<i>Pseudomonas</i>	0.117	0.097	0.097	0.091	0.005	0.021	0.024	0.227
<i>Moorella</i>	0.092	0.098	0.100	0.100	0.004	0.285	0.490	0.488

<i>Spirosoma</i>	0.105	0.090	0.099	0.095	0.003	0.792	0.009	0.129
<i>Anaerococcus</i>	0.093	0.082	0.103	0.108	0.005	0.001	0.487	0.109

55 Sequence data were submitted for bioinformatic analysis at MG RAST ([www.mg-rast.org](http://www.mg-rast.org)) before April 2020 when the family *Lactobacillaceae* was  
 56 reclassified (1). Therefore, the genus name *Lactobacillus* presented here includes the new genera *Lactobacillus*, *Lacticaseibacillus*,  
 57 *Lactiplantibacillus*, *Levilactobacillus*, *Ligilactobacillus* and *Limosilactobacillus*. Values are presented as least square means  $\pm$  SEM (n = 8 pigs per  
 58 dietary treatment). <sup>a,b</sup>Different superscripts within a row indicate significant difference ( $P \leq 0.05$ ). Con, control; LA, lactic acid-treated cereals.

59

## 60 Reference

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65 **TABLE S4** Bacterial abundances at amplicon sequence variant (ASV) level in cecal digesta of pigs fed diets with or without phytase and  
 66 lactic acid treatment of cereals

Treatment of cereal grains	No phytase		Phytase		SEM	P-value		
	Control	LA	Control	LA		Phytase	LA	Phytase x LA
<i>Prevotella copri</i>	10.11	13.67	7.92	8.62	1.148	0.004	0.075	0.225
<i>Prevotella ruminicola</i>	6.19	5.66	5.44	5.21	0.303	0.057	0.221	0.620
<i>Prevotella melaninogenica</i>	4.52	5.17	3.75	3.98	0.321	0.005	0.180	0.520
<i>Eubacterium rectale</i>	2.37	2.76	3.12	2.80	0.280	0.167	0.896	0.215
<i>Lactobacillus johnsonii</i>	1.58	2.06	0.98	2.15	0.652	0.700	0.218	0.601
<i>Faecalibacterium prausnitzii</i>	1.42	1.53	1.89	1.84	0.227	0.099	0.894	0.752
<i>Mitsuokella multacida</i>	1.44	2.33	1.13	1.41	0.180	0.002	0.003	0.103
<i>Prevotella buccae</i>	1.67	1.62	1.45	1.49	0.093	0.074	0.997	0.605
<i>Bacteroides vulgatus</i>	1.66	1.50	1.53	1.47	0.064	0.216	0.101	0.422
<i>Bacteroides fragilis</i>	1.61	1.40	1.50	1.42	0.057	0.464	0.017	0.284
<i>Lactobacillus acidophilus</i>	0.98	1.23	1.84	0.99	0.353	0.380	0.404	0.133
<i>Lactobacillus amylovorus</i>	0.98	1.22	1.87	0.93	0.364	0.420	0.347	0.116
<i>Prevotella oris</i>	1.19	1.16	1.03	1.08	0.065	0.082	0.846	0.603
<i>Prevotella bryantii</i>	1.11	1.05	0.98	1.02	0.054	0.137	0.784	0.376
<i>Bacteroides thetaiotaomicron</i>	1.11	1.01	1.01	0.98	0.041	0.120	0.139	0.339
<i>Eubacterium eligens</i>	0.98	0.84	1.19	1.06	0.098	0.037	0.187	0.998
<i>Bacteroides sp. 1_1_6</i>	1.11 a	0.91 b	1.01 a	0.99 b	0.044	0.813	0.018	0.044
<i>Limosilactobacillus reuteri</i> <sup>c</sup>	0.61	1.19	0.95	1.22	0.253	0.463	0.110	0.539
<i>Bacteroides sp. 4_3_47FAA</i>	1.02	0.88	0.97	0.89	0.039	0.532	0.008	0.402
<i>Prevotella bergenensis</i>	0.94	1.04	0.84	0.90	0.059	0.046	0.209	0.708
<i>Clostridium phytofermentans</i>	0.80	0.62	0.91	0.88	0.083	0.033	0.204	0.365
<i>Prevotella tannerae</i>	0.88	0.72	0.84	0.74	0.074	0.875	0.084	0.681
<i>Roseburia inulinivorans</i>	0.63	0.60	0.87	0.80	0.086	0.018	0.586	0.802
<i>Clostridium saccharolyticum</i>	0.71	0.57	0.79	0.79	0.057	0.014	0.261	0.213
<i>Ruminococcus albus</i>	0.75	0.54	0.76	0.73	0.067	0.147	0.093	0.197
<i>Prevotella buccalis</i>	0.74	0.66	0.67	0.68	0.035	0.417	0.300	0.211

<i>Ruminococcus</i> sp. 5_1_39BFAA	0.59	0.62	0.62	0.87	0.068	0.055	0.055	0.119
<i>Roseburia intestinalis</i>	0.61	0.56	0.78	0.73	0.065	0.016	0.457	0.971
<i>Parabacteroides distasonis</i>	0.68	0.55	0.68	0.65	0.032	0.134	0.018	0.092
<i>Prevotella timonensis</i>	0.67	0.61	0.61	0.62	0.031	0.399	0.410	0.242
<i>Clostridium botulinum</i>	0.58 b	0.45 b	0.59 b	0.81 a	0.071	0.017	0.500	0.022
<i>Clostridium difficile</i>	0.57 b	0.44 b	0.50 b	0.91 a	0.101	0.060	0.164	0.013
<i>Prevotella oralis</i>	0.64	0.58	0.58	0.59	0.033	0.395	0.399	0.297
<i>Selenomonas sputigena</i>	0.53	0.64	0.65	0.55	0.071	0.852	0.991	0.160
<i>Prevotella</i> sp. oral taxon 317	0.58	0.49	0.53	0.51	0.026	0.636	0.047	0.189
<i>Prevotella veroralis</i>	0.55	0.56	0.45	0.48	0.038	0.031	0.658	0.762
<i>Prevotella marshii</i>	0.56	0.43	0.54	0.50	0.025	0.408	0.003	0.077
<i>Prevotella</i> sp. oral taxon 472	0.54	0.46	0.51	0.48	0.023	0.804	0.024	0.220
<i>Lactobacillus helveticus</i>	0.38	0.47	0.70	0.39	0.133	0.393	0.423	0.145
<i>Clostridium beijerinckii</i>	0.45 b	0.35 b	0.45 b	0.64 a	0.058	0.019	0.408	0.023
<i>Bacteroides capillosus</i>	0.51	0.33	0.53	0.53	0.047	0.034	0.059	0.062
<i>Lactobacillus crispatus</i>	0.33	0.42	0.60	0.47	0.130	0.246	0.888	0.412
<i>Ruminococcus obeum</i>	0.43	0.39	0.43	0.56	0.049	0.085	0.373	0.101
<i>Lactobacillus gasseri</i>	0.39	0.51	0.31	0.53	0.127	0.794	0.186	0.686
<i>Clostridium thermocellum</i>	0.42	0.36	0.47	0.47	0.030	0.008	0.338	0.334
<i>Bacteroides dorei</i>	0.48	0.39	0.44	0.41	0.017	0.627	0.003	0.101
<i>Prevotella bivia</i>	0.46	0.44	0.40	0.41	0.024	0.059	0.983	0.545
<i>Lactobacillus ultunensis</i>	0.32	0.40	0.63	0.31	0.123	0.405	0.344	0.113
<i>Bacteroides plebeius</i>	0.45	0.35	0.46	0.40	0.019	0.150	0.001	0.290
<i>Bacteroides ovatus</i>	0.43	0.39	0.39	0.40	0.018	0.430	0.378	0.132
<i>Clostridium perfringens</i>	0.39 b	0.28 b	0.37 b	0.54 a	0.056	0.053	0.573	0.024
<i>Prevotella disiens</i>	0.44	0.39	0.36	0.37	0.025	0.049	0.553	0.312
<i>Bacteroides</i> sp. 2_1_16	0.42	0.36	0.40	0.37	0.016	0.838	0.010	0.549
<i>Bacteroides coprocola</i>	0.42	0.33	0.42	0.37	0.020	0.412	0.002	0.289
<i>Prevotella</i> sp. oral taxon 299	0.42	0.34	0.39	0.38	0.018	0.837	0.016	0.129
<i>Bacteroides</i> sp. 20_3	0.36	0.26	0.37	0.35	0.026	0.081	0.021	0.124
<i>Lactobacillus delbrueckii</i>	0.28	0.35	0.25	0.36	0.099	0.939	0.399	0.811
<i>Eubacterium hallii</i>	0.32	0.28	0.33	0.30	0.028	0.622	0.214	0.826
<i>Bacteroides coprophilus</i>	0.32	0.26	0.31	0.28	0.013	0.471	0.002	0.152

<i>Bacteroides pectinophilus</i>	0.26	0.26	0.33	0.29	0.025	0.053	0.492	0.391
<i>Parabacteroides sp. D13</i>	0.30	0.23	0.30	0.29	0.020	0.134	0.032	0.108
<i>Bacteroides cellulosilyticus</i>	0.28	0.24	0.28	0.26	0.011	0.329	0.015	0.246
<i>Clostridium bartlettii</i>	0.23 b	0.17 b	0.19 b	0.42 a	0.054	0.057	0.128	0.010
<i>Clostridium nexile</i>	0.239	0.196	0.283	0.268	0.024	0.022	0.239	0.558
<i>Eubacterium limosum</i>	0.230	0.196	0.254	0.262	0.016	0.011	0.436	0.222
<i>Clostridium sp. M62/1</i>	0.237	0.191	0.262	0.249	0.019	0.035	0.123	0.393
<i>Ruminococcus flavefaciens</i>	0.285	0.167	0.256	0.231	0.031	0.574	0.030	0.144
<i>Clostridium kluyveri</i>	0.219	0.201	0.242	0.259	0.014	0.010	0.965	0.231
<i>Bacteroides stercoris</i>	0.246	0.210	0.234	0.224	0.010	0.921	0.034	0.202
<i>Clostridium acetobutylicum</i>	0.214	0.196	0.233	0.261	0.015	0.008	0.741	0.133
<i>Bacteroides intestinalis</i>	0.247 a	0.192 b	0.236 a	0.221 a	0.010	0.379	0.002	0.047
<i>Clostridium cellulolyticum</i>	0.212	0.178	0.239	0.242	0.016	0.008	0.329	0.251
<i>Selenomonas flueggei</i>	0.191	0.247	0.217	0.200	0.020	0.605	0.354	0.083
<i>Bacteroides uniformis</i>	0.230	0.186	0.223	0.209	0.010	0.423	0.006	0.118
<i>Clostridium sp. L2-50</i>	0.211	0.185	0.225	0.209	0.022	0.403	0.346	0.824
<i>Clostridium leptum</i>	0.204	0.173	0.205	0.216	0.023	0.358	0.673	0.366
<i>Selenomonas noxia</i>	0.179	0.226	0.205	0.187	0.020	0.730	0.475	0.112
<i>Ruminococcus lactaris</i>	0.191	0.167	0.228	0.208	0.018	0.040	0.241	0.916
<i>Clostridium bolteae</i>	0.203	0.153	0.221	0.216	0.016	0.017	0.098	0.166
<i>Bacteroides sp. 2_1_7</i>	0.212 a	0.160 b	0.201 a	0.199 a	0.011	0.250	0.028	0.037
<i>Clostridium hathewayi</i>	0.193	0.141	0.224	0.209	0.018	0.012	0.078	0.312
<i>Eubacterium siraeum</i>	0.188	0.118	0.206	0.240	0.043	0.118	0.682	0.247
<i>Clostridium scindens</i>	0.188	0.144	0.207	0.206	0.018	0.030	0.205	0.230
<i>Clostridium asparagiforme</i>	0.188	0.140	0.210	0.202	0.015	0.010	0.075	0.199
<i>Ruminococcus gnavus</i>	0.177	0.146	0.213	0.194	0.020	0.049	0.237	0.796
<i>Bacteroides eggerthii</i>	0.199	0.151	0.197	0.181	0.010	0.176	0.004	0.121
<i>Bacteroides sp. D2</i>	0.193	0.169	0.184	0.179	0.008	0.940	0.059	0.225
<i>Clostridium cellulovorans</i>	0.174	0.148	0.191	0.209	0.015	0.013	0.806	0.142
<i>Clostridium sp. SS2/1</i>	0.174	0.134	0.184	0.198	0.018	0.050	0.464	0.137
<i>Parabacteroides merdae</i>	0.187	0.124	0.193	0.174	0.012	0.031	0.003	0.092
<i>Parabacteroides johnsonii</i>	0.186	0.123	0.190	0.174	0.013	0.043	0.006	0.082
<i>Bacteroides sp. 2_2_4</i>	0.176	0.150	0.170	0.166	0.007	0.467	0.027	0.118

<i>Eubacterium ventriosum</i>	0.158	0.129	0.185	0.177	0.017	0.041	0.293	0.545
<i>Bacteroides finegoldii</i>	0.177	0.150	0.161	0.161	0.007	0.719	0.087	0.076
<i>Clostridium novyi</i>	0.152	0.137	0.166	0.190	0.011	0.007	0.688	0.100
<i>Prevotella amnii</i>	0.178	0.166	0.149	0.143	0.009	0.009	0.362	0.736
<i>Bacteroides sp. D20</i>	0.166	0.135	0.161	0.151	0.006	0.436	0.004	0.117
<i>Eubacterium cellulosolvens</i>	0.138	0.131	0.152	0.172	0.017	0.120	0.704	0.427
<i>Clostridium sp. 7_2_43FAA</i>	0.126 b	0.078 b	0.112 b	0.264 a	0.047	0.077	0.276	0.041
<i>Selenomonas sp. oral taxon 137</i>	0.126	0.160	0.146	0.132	0.013	0.758	0.453	0.092

67 Values are presented as least square means  $\pm$  SEM (n = 8 pigs per dietary treatment). <sup>a,b</sup>Different superscripts within a row indicate  
 68 significant difference ( $P \leq 0.05$ ). Con, control; LA, lactic acid-treated cereals. <sup>c</sup> Due to the reclassification of the family *Lactobacillaceae*  
 69 (1), *Lactobacillus reuteri* was reclassified as *Limosilactobacillus reuteri*.

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72 **TABLE S5** Permutational multivariate analysis of variance (PERMANOVA) results for KEGG  
73 orthology (KO) functions (relative abundance > 0.01% of all reads) in cecal digesta of pigs fed  
74 diets with or without phytase and lactic acid treatment of cereals

Source of Variation	df	SS	R <sup>2</sup>	F	P-value
Diet	3	0.008	0.099	1.036	0.411 <sup>75</sup> <sup>76</sup>
Phytase	1	0.002	0.030	0.941	0.459 <sup>77</sup> <sup>78</sup>
Lactic acid	1	0.004	0.049	1.509	0.156 <sup>79</sup> <sup>79</sup>
Phytase : Lactic acid	1	0.002	0.021	0.659	0.679 <sup>80</sup> <sup>80</sup>
Residual	28	0.073	0.900		81
Total	31	0.081	1.000		82

84 The analysis based on pairwise distance of a multivariate data set and values were obtained using  
85 type III sums of squares with 999 permutations of residuals. Significant difference was declared  
86 at  $P \leq 0.05$ . df, degrees of freedom; SS, sum of squares.

87 **TABLE S6** KEGG orthology (KO) functions related to ‘carbohydrate metabolism’ pathways in cecal digesta of pigs fed diets with or  
 88 without phytase and lactic acid treatment of cereals

Treatment of grains	No phytase		Phytase		SEM	P-value		
	CON	LA	CON	LA		Phytase	LA	Phytase × LA
<b>00010 Glycolysis / Gluconeogenesis [PATH:ko00010]</b>								
E4.1.1.49, pckA; phosphoenolpyruvate carboxykinase (ATP) [EC:4.1.1.49]	0.33	0.33	0.31	0.31	0.0086	0.061	0.873	0.864
PGK, pgk; phosphoglycerate kinase [EC:2.7.2.3]	0.27	0.26	0.27	0.27	0.0031	0.052	0.048	0.924
GPI, pgi; glucose-6-phosphate isomerase [EC:5.3.1.9]	0.24	0.23	0.24	0.24	0.0042	0.076	0.100	0.170
pfkA, PFK; 6-phosphofructokinase 1 [EC:2.7.1.11]	0.23	0.21	0.25	0.26	0.0091	0.004	0.439	0.175
E3.2.1.86B, bglA; 6-phospho-beta-glucosidase [EC:3.2.1.86]	0.16	0.22	0.21	0.21	0.0190	0.235	0.097	0.197
FBA, fbaA; fructose-bisphosphate aldolase, class II [EC:4.1.2.13]	0.18	0.17	0.18	0.18	0.0039	0.054	0.097	0.307
fbp3; fructose-1,6-bisphosphatase III [EC:3.1.3.11]	0.16	0.16	0.16	0.16	0.0047	0.948	0.792	0.696
E5.1.3.3, galM; aldose 1-epimerase [EC:5.1.3.3]	0.15	0.16	0.16	0.16	0.0037	0.783	0.881	0.667
TPI, tpiA; triosephosphate isomerase (TIM) [EC:5.3.1.1]	0.12	0.12	0.13	0.13	0.0040	0.006	0.333	0.861
ACSS, acs; acetyl-CoA synthetase [EC:6.2.1.1]	0.12	0.11	0.10	0.11	0.0054	0.062	0.918	0.236
porA; pyruvate ferredoxin oxidoreductase, alpha subunit [EC:1.2.7.1]	0.032	0.022	0.035	0.032	0.0036	0.091	0.116	0.360
porB; pyruvate ferredoxin oxidoreductase, beta subunit [EC:1.2.7.1]	0.026	0.018	0.026	0.025	0.0027	0.254	0.094	0.182
glpX; fructose-1,6-bisphosphatase II [EC:3.1.3.11]	0.015	0.017	0.011	0.013	0.0014	0.009	0.129	0.928
porG; pyruvate ferredoxin oxidoreductase, gamma subunit [EC:1.2.7.1]	0.012	0.008	0.014	0.013	0.0015	0.030	0.096	0.322
E1.2.7.5, aor; aldehyde:ferredoxin oxidoreductase [EC:1.2.7.5]	0.0086	0.0050	0.0094	0.0083	0.0014	0.155	0.105	0.378
E3.2.1.86A, celF; 6-phospho-beta-glucosidase [EC:3.2.1.86]	0.0046	0.0035	0.0043	0.0051	0.0005	0.217	0.802	0.085
FBP, fbp; fructose-1,6-bisphosphatase I [EC:3.1.3.11]	0.0048	0.0043	0.0028	0.0012	0.0007	0.001	0.162	0.467
porD; pyruvate ferredoxin oxidoreductase, delta subunit [EC:1.2.7.1]	0.0026	0.0025	0.0032	0.0028	0.0003	0.087	0.382	0.559
agp; glucose-1-phosphatase [EC:3.1.3.10]	0.0021	0.0027	0.0026	0.0029	0.0004	0.318	0.246	0.712
E1.1.1.2, adh; alcohol dehydrogenase (NADP+) [EC:1.1.1.2]	0.0033	0.0033	0.0020	0.0014	0.0005	0.002	0.566	0.464
E1.2.1.9, gapN; glyceraldehyde-3-phosphate dehydrogenase (NADP) [EC:1.2.1.9]	0.0030	0.0016	0.0029	0.0025	0.0005	0.445	0.117	0.419
E6.2.1.13; acetyl-CoA synthetase (ADP-forming) [EC:6.2.1.13]	0.0021	0.0028	0.0024	0.0022	0.0004	0.737	0.598	0.248
ppgK; polyphosphate glucokinase [EC:2.7.1.63]	0.0024	0.0023	0.0012	0.0006	0.0004	0.004	0.419	0.630

**00020 Citrate cycle (TCA cycle) [PATH:ko00020]**

korA; 2-oxoglutarate ferredoxin oxidoreductase subunit alpha [EC:1.2.7.3]	0.20	0.17	0.18	0.17	0.0077	0.255	0.061	0.260
CS, gltA; citrate synthase [EC:2.3.3.1]	0.15	0.15	0.14	0.14	0.0073	0.385	0.795	0.759
E4.2.1.2A, fumA, fumB; fumarate hydratase, class I [EC:4.2.1.2]	0.14	0.13	0.12	0.11	0.0069	0.015	0.438	0.906
korB; 2-oxoglutarate ferredoxin oxidoreductase subunit beta [EC:1.2.7.3]	0.13	0.12	0.12	0.12	0.0047	0.271	0.116	0.461
E6.4.1.1B, pycB; pyruvate carboxylase subunit B [EC:6.4.1.1]	0.12	0.11	0.11	0.10	0.0050	0.112	0.409	0.756
mdh; malate dehydrogenase [EC:1.1.1.37]	0.091	0.091	0.081	0.077	0.0037	0.005	0.579	0.616
sdhB; succinate dehydrogenase iron-sulfur subunit [EC:1.3.99.1]	0.078	0.079	0.074	0.068	0.0036	0.049	0.543	0.309
PC, pyc; pyruvate carboxylase [EC:6.4.1.1]	0.056	0.062	0.047	0.056	0.0032	0.032	0.030	0.659
E4.2.1.2AA, fumA; fumarate hydratase subunit alpha [EC:4.2.1.2]	0.053	0.050	0.058	0.059	0.0036	0.049	0.797	0.558
korG; 2-oxoglutarate ferredoxin oxidoreductase subunit gamma [EC:1.2.7.3]	0.049	0.052	0.045	0.044	0.0026	0.016	0.674	0.473
E4.2.1.2AB, fumB; fumarate hydratase subunit beta [EC:4.2.1.2]	0.037	0.036	0.040	0.042	0.0028	0.108	0.887	0.648
sdhC; succinate dehydrogenase cytochrome b556 subunit	0.038	0.038	0.035	0.034	0.0023	0.154	0.770	0.987
sucC; succinyl-CoA synthetase beta subunit [EC:6.2.1.5]	0.027	0.038	0.023	0.025	0.0026	0.002	0.018	0.113
IDH3; isocitrate dehydrogenase (NAD+) [EC:1.1.1.41]	0.021	0.023	0.024	0.026	0.0020	0.147	0.486	0.921
sucD; succinyl-CoA synthetase alpha subunit [EC:6.2.1.5]	0.023	0.031	0.019	0.021	0.0021	0.002	0.017	0.145
korD; 2-oxoglutarate ferredoxin oxidoreductase subunit delta [EC:1.2.7.3]	0.013	0.015	0.012	0.011	0.0008	0.009	1.000	0.187
acnB; aconitate hydratase 2 / 2-methylisocitrate dehydratase [EC:4.2.1.3 4.2.1.99]	0.0220	0.0172	0.0081	0.0037	0.0029	<0.001	0.118	0.948
E6.4.1.1A, pycA; pyruvate carboxylase subunit A [EC:6.4.1.1]	0.0090	0.0115	0.0092	0.0074	0.0012	0.120	0.788	0.084
LSC1; succinyl-CoA synthetase alpha subunit [EC:6.2.1.4 6.2.1.5]	0.0010	0.0013	0.0008	0.0009	0.0001	0.025	0.181	0.470

**00030 Pentose phosphate pathway [PATH:ko00030]**

E2.2.1.1, tktA, tktB; transketolase [EC:2.2.1.1]	0.36	0.33	0.35	0.35	0.0087	0.446	0.074	0.138
PRPS, prsA; ribose-phosphate pyrophosphokinase [EC:2.7.6.1]	0.19	0.18	0.19	0.19	0.0049	0.129	0.473	0.740
deoC, DERA; deoxyribose-phosphate aldolase [EC:4.1.2.4]	0.13	0.13	0.13	0.14	0.0032	0.770	0.047	0.657
kdgK; 2-dehydro-3-deoxygluconokinase [EC:2.7.1.45]	0.13	0.12	0.13	0.12	0.0047	0.530	0.104	0.575
rpe, RPE; ribulose-phosphate 3-epimerase [EC:5.1.3.1]	0.13	0.12	0.13	0.13	0.0029	0.324	0.733	0.332
deoB; phosphopentomutase [EC:5.4.2.7]	0.079	0.084	0.094	0.095	0.0040	0.004	0.421	0.596
rpiB; ribose 5-phosphate isomerase B [EC:5.3.1.6]	0.072	0.080	0.071	0.074	0.0022	0.140	0.022	0.306
E4.1.2.9; phosphoketolase [EC:4.1.2.9]	0.043	0.054	0.055	0.055	0.0064	0.307	0.361	0.428
E1.1.1.44, PGD, gnd; 6-phosphogluconate dehydrogenase [EC:1.1.1.44]	0.030	0.041	0.038	0.035	0.0048	0.853	0.410	0.131

E2.2.1.2, talA, talB; transaldolase [EC:2.2.1.2]	0.032	0.033	0.034	0.034	0.0015	0.372	0.731	0.970
rbsK, RBKS; ribokinase [EC:2.7.1.15]	0.028	0.036	0.034	0.034	0.0035	0.634	0.280	0.264
G6PD, zwf; glucose-6-phosphate 1-dehydrogenase [EC:1.1.1.49]	0.024	0.032	0.033	0.028	0.0051	0.629	0.781	0.211
E2.7.1.12, gntK, idnK; gluconokinase [EC:2.7.1.12]	0.022	0.033	0.031	0.028	0.0044	0.634	0.345	0.108
pgl; 6-phosphogluconolactonase [EC:3.1.1.31]	0.018	0.024	0.025	0.024	0.0028	0.206	0.425	0.166
rpiA; ribose 5-phosphate isomerase A [EC:5.3.1.6]	0.019	0.019	0.020	0.019	0.0023	0.945	0.996	0.756
hxlB; 6-phospho-3-hexuloisomerase [EC:5.3.1.27]	0.0010	0.0011	0.0006	0.0015	0.0002	0.957	0.053	0.137

#### 00052 Galactose metabolism [PATH:ko00052]

lacZ; beta-galactosidase [EC:3.2.1.23]	0.65	0.65	0.65	0.65	0.0229	0.968	0.977	0.907
E3.2.1.22B, galA, rafA; alpha-galactosidase [EC:3.2.1.22]	0.34	0.34	0.32	0.36	0.0116	0.703	0.070	0.190
gale, GALE; UDP-glucose 4-epimerase [EC:5.1.3.2]	0.29	0.28	0.30	0.30	0.0063	0.011	0.147	0.450
E3.2.1.20, malZ; alpha-glucosidase [EC:3.2.1.20]	0.21	0.23	0.23	0.23	0.0117	0.279	0.303	0.461
galK; galactokinase [EC:2.7.1.6]	0.16	0.16	0.17	0.17	0.0040	0.214	0.971	0.752
bgaB, lacA; beta-galactosidase [EC:3.2.1.23]	0.14	0.13	0.16	0.16	0.0069	0.004	0.562	0.428
galT, GALT; UDPglucose--hexose-1-phosphate uridylyltransferase [EC:2.7.7.12]	0.10	0.12	0.12	0.13	0.0070	0.057	0.181	0.542
E3.2.1.10; oligo-1,6-glucosidase [EC:3.2.1.10]	0.067	0.077	0.076	0.082	0.0044	0.142	0.079	0.712
E3.2.1.26, sacA; beta-fructofuranosidase [EC:3.2.1.26]	0.059	0.069	0.073	0.076	0.0044	0.025	0.135	0.445
E3.2.1.85, lacG; 6-phospho-beta-galactosidase [EC:3.2.1.85]	0.022	0.032	0.020	0.030	0.0037	0.519	0.012	0.975
lacC; tagatose 6-phosphate kinase [EC:2.7.1.144]	0.011	0.013	0.014	0.011	0.0019	0.733	0.738	0.213
melA; alpha-galactosidase [EC:3.2.1.22]	0.008	0.005	0.011	0.011	0.0018	0.024	0.340	0.509
gatY-kbaY; tagatose 1,6-diphosphate aldolase GatY/KbaY [EC:4.1.2.40]	0.0070	0.0110	0.0068	0.0084	0.0010	0.164	0.008	0.249
E5.3.1.26, lacA, lacB; galactose-6-phosphate isomerase [EC:5.3.1.26]	0.0048	0.0093	0.0050	0.0070	0.0016	0.522	0.056	0.466
ebgA; evolved beta-galactosidase subunit alpha [EC:3.2.1.23]	0.0064	0.0049	0.0060	0.0064	0.0006	0.388	0.323	0.130
lacD; tagatose 1,6-diphosphate aldolase [EC:4.1.2.40]	0.0060	0.0069	0.0038	0.0066	0.0022	0.567	0.420	0.675
dgoD; galactonate dehydratase [EC:4.2.1.6]	0.0045 a	0.0032 b	0.0041 ab	0.0052 a	0.0005	0.151	0.873	0.039

#### 00500 Starch and sucrose metabolism [PATH:ko00500]

E2.4.1.1, glgP, PYG; starch phosphorylase [EC:2.4.1.1]	0.41	0.39	0.43	0.42	0.0123	0.039	0.130	0.637
glgB; 1,4-alpha-glucan branching enzyme [EC:2.4.1.18]	0.30	0.28	0.30	0.29	0.0068	0.367	0.138	0.533
malQ; 4-alpha-glucanotransferase [EC:2.4.1.25]	0.27	0.26	0.27	0.27	0.0068	0.193	0.339	0.265

glgC; glucose-1-phosphate adenylyltransferase [EC:2.7.7.27]	0.17	0.17	0.19	0.18	0.0086	0.142	0.645	0.270
E2.4.1.21, glgA; starch synthase [EC:2.4.1.21]	0.16	0.16	0.17	0.17	0.0062	0.234	0.627	0.550
E3.2.1.1A; alpha-amylase [EC:3.2.1.1]	0.092	0.092	0.084	0.086	0.0053	0.162	0.858	0.833
E2.4.1.8, mapA; maltose phosphorylase [EC:2.4.1.8]	0.048	0.066	0.057	0.059	0.0093	0.877	0.312	0.387
E3.2.1.4; endoglucanase [EC:3.2.1.4]	0.059	0.048	0.058	0.055	0.0030	0.351	0.031	0.143
rfbF; glucose-1-phosphate cytidylyltransferase [EC:2.7.7.33]	0.060	0.052	0.049	0.047	0.0038	0.054	0.171	0.462
E2.4.1.7; sucrose phosphorylase [EC:2.4.1.7]	0.035	0.049	0.053	0.052	0.0057	0.088	0.286	0.227
xynB; xylan 1,4-beta-xylosidase [EC:3.2.1.37]	0.033	0.034	0.036	0.034	0.0027	0.567	0.654	0.527
E3.2.1.1, amyA, malS; alpha-amylase [EC:3.2.1.1]	0.028	0.028	0.027	0.033	0.0023	0.399	0.185	0.232
E3.2.1.122, glvA; maltose-6'-phosphate glucosidase [EC:3.2.1.122]	0.026	0.028	0.031	0.030	0.0041	0.458	0.804	0.668
treC; trehalose-6-phosphate hydrolase [EC:3.2.1.93]	0.024	0.032	0.024	0.031	0.0029	0.821	0.012	0.906
treS; maltose alpha-D-glucosyltransferase/ alpha-amylase [EC:5.4.99.16 3.2.1.1]	0.017	0.019	0.020	0.019	0.0013	0.202	0.504	0.285
pgmB; beta-phosphoglucomutase [EC:5.4.2.6]	0.015	0.018	0.019	0.019	0.0023	0.348	0.456	0.552
E2.4.1.20; cellobiose phosphorylase [EC:2.4.1.20]	0.015	0.012	0.014	0.015	0.0017	0.748	0.604	0.208
E2.4.1.4; amylosucrase [EC:2.4.1.4]	0.011	0.012	0.012	0.014	0.0012	0.233	0.324	0.562
E3.2.1.54; cyclomaltodextrinase [EC:3.2.1.54]	0.0022	0.0022	0.0031	0.0041	0.0004	0.001	0.172	0.193
E3.2.1.91; cellulose 1,4-beta-cellulosidase [EC:3.2.1.91]	0.0026	0.0015	0.0015	0.0018	0.0003	0.165	0.190	0.037
ENPP1_3; ectonucleotide pyrophosphatase/phosphodiesterase family member 1/3 [EC:3.1.4.1 3.6.1.9]	0.0011	0.0013	0.0008	0.0010	0.0002	0.165	0.529	0.928

#### 00520 Amino sugar and nucleotide sugar metabolism [PATH:ko00520]

murA; UDP-N-acetylglucosamine 1-carboxyvinyltransferase [EC:2.5.1.7]	0.28	0.28	0.29	0.30	0.0065	0.010	0.840	0.322
wecB; UDP-N-acetylglucosamine 2-epimerase [EC:5.1.3.14]	0.20	0.20	0.21	0.19	0.0075	0.927	0.306	0.376
nagB, GNPDA; glucosamine-6-phosphate deaminase [EC:3.5.99.6]	0.19	0.21	0.17	0.19	0.0115	0.057	0.101	0.674
glmM; phosphoglucosamine mutase [EC:5.4.2.10]	0.15	0.15	0.16	0.16	0.0062	0.061	1.000	0.783
murB; UDP-N-acetylmuramate dehydrogenase [EC:1.1.1.158]	0.11	0.12	0.12	0.12	0.0020	0.722	0.214	0.261
npdA; NAD-dependent deacetylase [EC:3.5.1.-]	0.11	0.11	0.12	0.12	0.0024	0.192	0.996	0.377
E3.2.1.55, abfA; alpha-N-arabinofuranosidase [EC:3.2.1.55]	0.11	0.10	0.11	0.11	0.0047	0.484	0.797	0.423
glmU; bifunctional UDP-N-acetylglucosamine pyrophosphorylase / Glucosamine-1-phosphate N-acetyltransferase [EC:2.7.7.23 2.3.1.157]	0.10	0.11	0.10	0.10	0.0050	0.768	0.306	0.658
E3.5.1.25, nagA, AMDHD2; N-acetylglucosamine-6-phosphate deacetylase [EC:3.5.1.25]	0.078	0.079	0.084	0.082	0.0033	0.241	0.924	0.623

E2.5.1.56, neuB; N-acetylneuraminate synthase [EC:2.5.1.56]	0.056	0.055	0.068	0.065	0.0077	0.183	0.792	0.925
murQ; N-acetylmuramic acid 6-phosphate etherase [EC:4.2.1.126]	0.050	0.065	0.046	0.053	0.0037	0.032	0.008	0.314
wbpO; UDP-N-acetyl-D-galactosamine dehydrogenase [EC:1.1.1.-]	0.049	0.056	0.054	0.040	0.0053	0.323	0.540	0.070
rfbG; CDP-glucose 4,6-dehydratase [EC:4.2.1.45]	0.055	0.050	0.044	0.045	0.0047	0.106	0.779	0.535
E4.1.3.3, nanA, NPL; N-acetylneuraminate lyase [EC:4.1.3.3]	0.038	0.037	0.041	0.039	0.0025	0.394	0.651	0.861
E3.2.1.52, nagZ; beta-N-acetylhexosaminidase [EC:3.2.1.52]	0.036	0.036	0.039	0.041	0.0020	0.061	0.637	0.650
rfbH; CDP-6-deoxy-D-xylo-4-hexulose-3-dehydratase	0.043	0.031	0.040	0.034	0.0034	0.977	0.016	0.369
RENBP; N-acylglucosamine 2-epimerase [EC:5.1.3.8]	0.030	0.031	0.031	0.029	0.0020	0.985	0.692	0.622
E2.7.7.43, neuA, CMAS; N-acetylneuraminate cytidyltransferase [EC:2.7.7.43]	0.020	0.019	0.022	0.023	0.0036	0.356	0.969	0.882
UAP1; UDP-N-acetylglucosamine pyrophosphorylase [EC:2.7.7.23]	0.018	0.018	0.021	0.019	0.0022	0.327	0.666	0.695
nanE; N-acylglucosamine-6-phosphate 2-epimerase [EC:5.1.3.9]	0.0098	0.0096	0.0103	0.0116	0.0009	0.198	0.559	0.436
wecC; UDP-N-acetyl-D-mannosaminuronic acid dehydrogenase [EC:1.1.1.336]	0.0068	0.0059	0.0072	0.0089	0.0009	0.061	0.675	0.132
rfbE; CDP-paratose 2-epimerase [EC:5.1.3.10]	0.0010	0.0015	0.0006	0.0010	0.0005	0.304	0.317	0.968

#### 00620 Pyruvate metabolism [PATH:ko00620]

ppdK; pyruvate,orthophosphate dikinase [EC:2.7.9.1]	0.39 a	0.35 b	0.37 ab	0.39 a	0.0098	0.181	0.257	0.019
E2.3.1.54, pflD; formate C-acetyltransferase [EC:2.3.1.54]	0.24	0.27	0.26	0.29	0.0081	0.042	0.004	0.707
ackA; acetate kinase [EC:2.7.2.1]	0.24	0.23	0.27	0.26	0.0097	0.003	0.177	0.666
E2.3.1.8, pta; phosphate acetyltransferase [EC:2.3.1.8]	0.15	0.15	0.15	0.15	0.0044	0.803	0.759	0.627
E1.1.1.40, maeB; malate dehydrogenase (oxaloacetate-decarboxylating)(NADP+) [EC:1.1.1.40]	0.15	0.14	0.13	0.13	0.0080	0.042	0.416	0.618
accC; acetyl-CoA carboxylase, biotin carboxylase subunit [EC:6.4.1.2 6.3.4.14]	0.074	0.077	0.080	0.077	0.0043	0.458	0.912	0.532
fucO; lactaldehyde reductase [EC:1.1.1.77]	0.074	0.074	0.082	0.076	0.0050	0.360	0.611	0.574
ldhA; D-lactate dehydrogenase [EC:1.1.1.28]	0.064	0.070	0.069	0.075	0.0055	0.341	0.254	0.955
ppc; phosphoenolpyruvate carboxylase [EC:4.1.1.31]	0.046	0.054	0.046	0.047	0.0071	0.670	0.523	0.655
accA; acetyl-CoA carboxylase carboxyl transferase subunit alpha [EC:6.4.1.2]	0.048	0.044	0.046	0.046	0.0020	0.874	0.339	0.410
accD; acetyl-CoA carboxylase carboxyl transferase subunit beta [EC:6.4.1.2]	0.038	0.037	0.039	0.041	0.0027	0.394	0.782	0.474
E3.1.2.1, ACH1; acetyl-CoA hydrolase [EC:3.1.2.1]	0.037	0.045	0.035	0.033	0.0024	0.008	0.203	0.078
E1.2.3.3, poxL; pyruvate oxidase [EC:1.2.3.3]	0.031	0.037	0.038	0.032	0.0061	0.868	0.895	0.335
pps, ppsA; pyruvate, water dikinase [EC:2.7.9.2]	0.028	0.037	0.024	0.032	0.0029	0.150	0.010	0.818
E4.2.3.3, mgsA; methylglyoxal synthase [EC:4.2.3.3]	0.026	0.024	0.030	0.030	0.0017	0.008	0.536	0.650

accB, bccP; acetyl-CoA carboxylase biotin carboxyl carrier protein	0.012	0.011	0.011	0.012	0.0008	0.610	0.987	0.276
aldA; lactaldehyde dehydrogenase / glycolaldehyde dehydrogenase [EC:1.2.1.22 1.2.1.21]	0.0091	0.0036	0.0088	0.0144	0.0028	0.068	1.000	0.057
acyP; acylphosphatase [EC:3.6.1.7]	0.0077	0.0103	0.0079	0.0089	0.0012	0.609	0.138	0.519
pct; propionate CoA-transferase [EC:2.8.3.1]	0.0068	0.0103	0.0061	0.0071	0.0010	0.063	0.029	0.211
E3.1.2.6, gloB; hydroxyacylglutathione hydrolase [EC:3.1.2.6]	0.0071	0.0053	0.0067	0.0067	0.0005	0.330	0.070	0.077
pta; phosphate acetyltransferase [EC:2.3.1.8]	0.0090	0.0071	0.0046	0.0029	0.0010	<0.001	0.100	0.920
mqo; malate dehydrogenase (quinone) [EC:1.1.5.4]	0.0046	0.0058	0.0031	0.0010	0.0013	0.019	0.711	0.208
ACAC; acetyl-CoA carboxylase / biotin carboxylase [EC:6.4.1.2 6.3.4.14]	0.0012 b	0.0007 b	0.0009 b	0.0022 a	0.0004	0.148	0.350	0.022
E1.2.1.10; acetaldehyde dehydrogenase (acetylating) [EC:1.2.1.10]	0.0011	0.0008	0.0016	0.0012	0.0002	0.018	0.030	0.794

#### 00640 Propanoate metabolism [PATH:ko00640]

mmdA; methylmalonyl-CoA decarboxylase alpha chain [EC:4.1.1.41]	0.012	0.011	0.010	0.012	0.0006	0.769	0.976	0.036
E2.3.3.5, prpC; 2-methylcitrate synthase [EC:2.3.3.5]	0.0098	0.0101	0.0117	0.0094	0.0013	0.664	0.427	0.341
mmdb; methylmalonyl-CoA decarboxylase beta chain [EC:4.1.1.41]	0.0073	0.0071	0.0072	0.0070	0.0004	0.798	0.600	0.988
E4.2.1.79, prpD; 2-methylcitrate dehydratase [EC:4.2.1.79]	0.0038	0.0055	0.0032	0.0006	0.0012	0.035	0.704	0.092
E4.1.3.30, prpB; methylisocitrate lyase [EC:4.1.3.30]	0.0028	0.0043	0.0020	0.0007	0.0009	0.025	0.913	0.132
E3.5.99.7; 1-aminocyclopropane-1-carboxylate deaminase [EC:3.5.99.7]	0.0016	0.0020	0.0013	0.0006	0.0004	0.027	0.788	0.138

#### 00650 Butanoate metabolism [PATH:ko00650]

E2.7.2.7, buk; butyrate kinase [EC:2.7.2.7]	0.077 a	0.059 b	0.074 a	0.076 a	0.0040	0.096	0.068	0.020
E4.1.1.70; glutaconyl-CoA decarboxylase [EC:4.1.1.70]	0.035	0.043	0.035	0.035	0.0020	0.060	0.063	0.083
ptb; phosphate butyryltransferase [EC:2.3.1.19]	0.038	0.029	0.038	0.041	0.0030	0.055	0.466	0.055
E4.1.1.5, alsD; acetolactate decarboxylase [EC:4.1.1.5]	0.0061	0.0076	0.0040	0.0075	0.0020	0.595	0.217	0.621
E4.2.1.-; [EC:4.2.1.-]	0.0048 a	0.0039 b	0.0033 b	0.0045 ab	0.0003	0.222	0.706	0.007
E1.1.1.30, bdh; 3-hydroxybutyrate dehydrogenase [EC:1.1.1.30]	0.0017 ab	0.0012 b	0.0014 ab	0.0018 a	0.0002	0.593	0.688	0.016
E1.3.1.44; trans-2-enoyl-CoA reductase (NAD+) [EC:1.3.1.44]	0.0017	0.0014	0.0014	0.0011	0.0002	0.173	0.315	0.915
E1.1.1.61; 4-hydroxybutyrate dehydrogenase [EC:1.1.1.61]	0.0015	0.0014	0.0011	0.0015	0.0001	0.279	0.241	0.109
gctB; glutaconate CoA-transferase, subunit B [EC:2.8.3.12]	0.0017 a	0.0012 ab	0.0010 b	0.0012 ab	0.0002	0.079	0.494	0.048

89 Values are presented as least square means  $\pm$  SEM. (n = 8 pigs per dietary treatment). <sup>a,b</sup> Different superscripts within a row indicate

90 significant difference ( $P \leq 0.05$ ). Con, control; LA, lactic acid-treated cereals.

91 **TABLE S7** Selected COGs (Clusters of Orthologous Groups) function in carbohydrate metabolism in cecal digesta of pigs fed diets with  
 92 or without phytase and lactic acid treatment of cereals

COG Function	No phytase		Phytase		SEM	P-value		
	Con	LA	Con	LA		Phytase	LA	Phytase×LA
Alpha-glucosidases, family 31 of glycosyl hydrolases	2.65	2.87	2.66	2.81	0.089	0.813	0.047	0.675
Sugar pentulose and hexulose kinases	2.57	2.77	2.53	2.47	0.084	0.050	0.404	0.136
Nucleoside-diphosphate-sugar epimerases	2.59	2.36	2.46	2.12	0.100	0.079	0.010	0.588
Phosphoenolpyruvate synthase pyruvate phosphate dikinase	2.42 a	2.08 b	2.30 a	2.32 a	0.085	0.461	0.066	0.048
Beta-glucosidase 6-phospho-beta-glucosidase beta-galactosidase	1.65	2.18	1.89	2.19	0.137	0.381	0.005	0.419
Alpha-galactosidase	1.78	2.00	1.72	1.95	0.072	0.444	0.004	0.919
Glucan phosphorylase	1.74	1.48	1.79	1.69	0.083	0.133	0.044	0.351
Predicted nucleoside-diphosphate sugar epimerases	1.70	1.38	1.82	1.57	0.098	0.125	0.008	0.690
Fucose permease	1.59	1.61	1.45	1.37	0.096	0.063	0.774	0.645
6-phosphofructokinase	1.56 a	1.32 b	1.52 a	1.56 a	0.065	0.144	0.143	0.039
3-phosphoglycerate kinase	1.49	1.41	1.48	1.40	0.031	0.840	0.016	0.926
1,4-alpha-glucan branching enzyme	1.22	1.08	1.22	1.13	0.052	0.633	0.036	0.558
ABC-type sugar transport systems, ATPase components	1.05	0.93	1.12	1.14	0.063	0.029	0.421	0.270
Cellobiose phosphorylase	1.07 a	0.95 b	1.03 a	1.11 b	0.045	0.165	0.615	0.036
6-phosphogluconolactonase	1.04	1.12	0.98	0.99	0.046	0.054	0.342	0.414
Glucosamine-6-phosphate isomerase deaminase								
Glucose-6-phosphate isomerase	1.03	0.94	1.03	1.01	0.027	0.240	0.045	0.224

Phosphotransferase system IIC components, glucose maltose N-acetylglucosamine-specific	0.87	1.19	0.90	1.06	0.108	0.679	0.034	0.470
Enolase	0.98	1.03	0.92	1.07	0.046	0.791	0.033	0.243
Dihydroxyacid dehydratase phosphogluconate dehydratase	1.02	0.93	0.97	0.90	0.036	0.233	0.037	0.855
Transketolase	1.06	0.90	0.98	0.84	0.051	0.194	0.008	0.757
4-alpha-glucanotransferase	0.97	0.85	1.01	0.92	0.050	0.334	0.046	0.755
Glyceraldehyde-3-phosphate dehydrogenase erythrose-4-phosphate dehydrogenase	0.86	0.79	0.85	0.79	0.024	0.724	0.016	0.936
Beta-galactosidase	0.81	0.71	0.92	0.85	0.037	0.003	0.023	0.650
Ribulose-5-phosphate 4-epimerase and related epimerases and aldolases	0.84	0.89	0.77	0.76	0.038	0.014	0.657	0.485
Pyruvate kinase	0.78	0.74	0.80	0.86	0.031	0.042	0.702	0.098
L-arabinose isomerase	0.75 b	0.90 a	0.79 b	0.72 b	0.040	0.093	0.323	0.011
ABC-type sugar transport systems, permease components	0.72	0.74	0.84	0.84	0.050	0.040	0.790	0.848
ABC-type sugar transport system, permease component	0.54	0.52	0.70	0.72	0.040	0.000	0.956	0.651
Uncharacterized protein conserved in bacteria	0.62	0.55	0.66	0.62	0.030	0.077	0.075	0.651
ABC-type sugar transport system, ATPase component	0.59	0.49	0.59	0.68	0.057	0.115	0.961	0.094
Phosphoenolpyruvate-protein kinase PTS system EI component in bacteria	0.49	0.59	0.55	0.64	0.035	0.126	0.012	0.971
Phosphopentomutase	0.45	0.53	0.56	0.56	0.030	0.021	0.197	0.164
Phosphotransferase system, mannose fructose N-acetylgalactosamine-specific component IID	0.45	0.61	0.46	0.52	0.057	0.436	0.068	0.391

Phosphotransferase system, mannose fructose N-acetylgalactosamine- specific component IIC	0.45	0.60	0.44	0.54	0.071	0.611	0.088	0.699
Predicted xylanase chitin deacetylase	0.49	0.37	0.50	0.55	0.044	0.034	0.494	0.056
Phosphomanno isomerase	0.48 a	0.43 b	0.47 a	0.49 a	0.012	0.038	0.195	0.027
5-keto 4-deoxyuronate isomerase	0.48	0.35	0.42	0.36	0.033	0.489	0.008	0.377
Transketolase, C-terminal subunit	0.38	0.34	0.36	0.39	0.018	0.423	0.852	0.063
Arabinogalactan endo-1,4-beta- galactosidase	0.34	0.35	0.38	0.38	0.019	0.080	0.721	0.781
Transcriptional regulators of sugar metabolism	0.31	0.44	0.31	0.37	0.029	0.263	0.004	0.281
TRAP-type C4-dicarboxylate transport system, large permease component	0.36	0.32	0.38	0.38	0.019	0.033	0.231	0.209
Transketolase, N-terminal subunit	0.37	0.33	0.35	0.39	0.020	0.284	0.883	0.072
Mannose-6-phosphate isomerase	0.35	0.39	0.29	0.25	0.030	0.003	0.950	0.185
Predicted phosphoglycerate mutase, AP superfamily	0.34	0.32	0.30	0.29	0.019	0.068	0.385	0.798
Pectin methylesterase	0.34	0.33	0.27	0.26	0.027	0.021	0.593	0.928
Arabinose efflux permease	0.27	0.39	0.26	0.27	0.032	0.044	0.052	0.086
Phosphoglycerate mutase 1	0.24	0.35	0.28	0.30	0.035	0.828	0.065	0.225
Endoglucanase Y	0.29	0.25	0.31	0.31	0.017	0.024	0.190	0.342
Phosphotransferase system IIA components	0.24	0.35	0.24	0.33	0.047	0.786	0.043	0.776
Endoglucanase	0.35 a	0.22 b	0.26 b	0.26 b	0.019	0.251	0.003	0.004
Cellulase M and related proteins	0.24	0.25	0.28	0.30	0.023	0.091	0.632	0.893
Beta-1,4-xylanase	0.30	0.27	0.23	0.22	0.017	0.001	0.333	0.715
2-keto-3-deoxy-6-phosphogluconate aldolase	0.25	0.18	0.24	0.24	0.016	0.187	0.026	0.060
Maltose-binding periplasmic	0.24	0.25	0.18	0.18	0.031	0.044	0.929	0.696

proteins domains								
Alpha-glucuronidase	0.21	0.22	0.19	0.18	0.015	0.082	0.885	0.538
Phosphotransferase system, mannose fructose-specific component IIA	0.17	0.26	0.13	0.22	0.044	0.343	0.067	0.981
Phosphoheptose isomerase	0.15 b	0.21 a	0.16 b	0.12 b	0.020	0.100	0.725	0.019
Xylose isomerase	0.18	0.14	0.19	0.10	0.019	0.457	0.002	0.232
TRAP-type C4-dicarboxylate transport system, periplasmic component	0.15	0.12	0.16	0.16	0.009	0.004	0.199	0.103
Beta-mannanase	0.16	0.11	0.14	0.14	0.013	0.974	0.045	0.134
ABC-type polysaccharide transport system, permease component	0.10	0.08	0.13	0.16	0.015	0.001	0.953	0.147
Pectate lyase	0.12	0.09	0.11	0.10	0.010	0.941	0.086	0.172
Neutral trehalase	0.090	0.062	0.102	0.109	0.015	0.061	0.495	0.261
Phosphotransferase system, HPr- related proteins	0.078	0.079	0.084	0.098	0.007	0.091	0.316	0.355
Citrate lyase beta subunit	0.085	0.094	0.064	0.090	0.008	0.112	0.026	0.267
Archaeal fructose-1,6-bisphosphatase and related enzymes of inositol monophosphatase family	0.098	0.067	0.083	0.068	0.007	0.330	0.004	0.309
Fructose-1,6-bisphosphatase sedoheptulose 1,7-bisphosphatase and related proteins	0.078	0.099	0.054	0.069	0.008	0.003	0.034	0.708
Phosphoglucomutase	0.095	0.088	0.052	0.043	0.010	0.000	0.445	0.924
Endo-beta-mannanase	0.080	0.058	0.066	0.068	0.007	0.737	0.132	0.083
Phosphotransferase system cellobiose- specific component IIB	0.056	0.066	0.056	0.092	0.008	0.110	0.009	0.117
TRAP-type C4-dicarboxylate transport system, small permease component	0.051	0.036	0.052	0.049	0.004	0.147	0.056	0.186

93 Values are presented as least square means  $\pm$  SEM. ( $n = 8$  pigs per dietary treatment). <sup>a,b</sup>Different superscripts within a row indicate  
94 significant difference ( $P \leq 0.05$ ). COGs (Clusters of Orthologous Groups); Con, control; LA, lactic acid-treated cereals. Only Clusters of  
95 Orthologous Group functions within the carbohydrate metabolism that were significantly ( $P \leq 0.05$ ) or tended ( $P \leq 0.10$ ) to be different  
96 among treatments are presented.

97 **TABLE S8** Permutational multivariate analysis of variance (PERMANOVA) results for KEGG  
98 orthology (KO) functions related to ‘carbohydrate metabolism’ (relative abundance > 0.01% of  
99 all reads) in cecal digesta of pigs fed diets with or without phytase and lactic acid treatment of  
100 cereals

Source of Variation	df	SS	R <sup>2</sup>	F	P-value
Diet	3	0.017	0.141	1.538	0.077
Phytase	1	0.009	0.076	2.481	0.031
Lactic acid	1	0.005	0.041	1.342	0.196
Phytase : Lactic acid	1	0.003	0.024	0.792	0.536
Residual	28	0.104	0.859		
Total	31	0.121	1.000		

101  
102 The analysis based on pairwise distance of a multivariate data set and values were obtained using  
103 type III sums of squares with 999 permutations of residuals. Significant difference was declared  
104 at  $P \leq 0.05$ . df, degrees of freedom; SS, sum of square.