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Physiological response to the weaning in two pig lines divergently selected for residual feed intake

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1 **i. Title**

2 Physiological response to the weaning in two pig lines divergently selected for residual feed
3 intake

4

5 **ii. Short running title**

6 Divergent selection for RFI and weaning

7

8 **iii. Authors**

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15

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21

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24 residual feed intake. 2016. 67th Annual Meeting of the European Federation of Animal
25 Science (EAAP), 29 August -2 September 2016, Belfast (UK), p 434 (abstract)

26

27 **vi. Abstract and Keywords**

28 Breeding efficient pigs is a way to reduce dietary costs and environmental waste. However,
29 optimisation of feed efficiency must not be linked to a decrease of the ability of animals to
30 cope with stress, such as the weaning. This study characterizes the response after weaning
31 of pigs from two lines divergently selected for residual feed intake (RFI) during growth.
32 Animals of the low (L) RFI line are more efficient than animals from the high (H) RFI line.
33 Thirty six piglets from each line, weaned at 28 days of age were individually housed and fed
34 a conventional dietary sequence. Their performance, behaviour, health and oxidative status,
35 immune and nutritional parameters were followed during three weeks. Daily feed intake and
36 growth rate of pigs from the LRFI line were 35% and 40% lower compared with HRFI
37 (P<0.001). Pigs from the LRFI-line had lower total tract apparent digestibility (−6% for OM)

38 and suffered more from undernutrition with a 167 and 55% higher plasmatic concentration of
39 NEFA and urea compared with HRFI ($P < 0.01$). In the first week after the weaning, they had
40 more diarrhoea, and had a higher inflammatory status with concentration of haptoglobin 52%
41 higher ($P < 0.001$). These piglets then seemed to adapt to the weaning conditions and to
42 recover during the second and third weeks. Both lines had similar zootechnical performance
43 and physiological characteristics at the end of the post-weaning period. To conclude, the
44 physiological responses to the weaning differed between lines. Pigs from the LRFI line,
45 selected for greater feed efficiency were more sensitive to the weaning stress. They were
46 also more resilient as they finally adapted to the new condition and recovered to show similar
47 performance results as pigs of the HRFI line.

48
49 Feed efficiency, pig, residual feed intake, selection, weaning

50

51 **vii. Main text**

52

53 **Introduction**

54 In commercial pig farms, the weaning is a critical period for pig due to an important sensibility
55 to digestive disorders. Then the weaning period has been often associated with the use of
56 antibiotics to prevent or to cure diarrhoea. In the worldwide context of reduction of the use of
57 antibiotics to limit bacterial resistance and preserve public health, there is an increased
58 interest to select piglets that have important ability to cope with the stress of weaning
59 (maternal separation, litter mixing, new, transportation, diet change, etc.). The immediate
60 consequences of the weaning stress are decreased voluntary feed intake, growth check and
61 increased sensitivity to enteric pathogens that compromise animal health and welfare (Molist
62 et al., 2014). These reflect physiological perturbations as altered gastrointestinal tract with
63 villus atrophy, compromised barrier function, reduced digestive and absorptive capacity,
64 microbiota dysbiosis (Pluske, Hampson, & Williams, 1997; Lallès, Bosi, Smidt, & Stokes,
65 2007; Lallès & Guillou, 2015), systemic inflammation, and oxidative stress (Zhu, Zhao, Chen,
66 & Xu, 2012; Buchet et al., 2017). This acute phase is usually followed by a recovery or
67 adaptive phase corresponding to an adaptation of the pig to its new rearing, feeding and
68 social conditions (Burrin & Stoll, 2003; Montagne et al., 2007). When focusing on the kinetic
69 of the piglet response after weaning, robustness at weaning could result 1) from a greater
70 resistance of piglets to the stressing factors, corresponding to a lower sensitivity during the
71 acute phase and/or 2) from a greater resilience to these factors, corresponding to a more
72 rapid recovery during the adaptive phase (Sauvant & Martin, 2010).

73 Improving feed efficiency is a major goal in pig production because of its relationships to
74 reduced feed costs and environmental impacts. Residual feed intake (RFI), defined as the

75 difference between observed feed intake and that predicted on the basis of requirements for
76 maintenance and growth, is a criteria to divergently select pigs for feed efficiency (Gilbert et
77 al., 2007). For a similar growth rate, pigs selected for low RFI (LRFI; more efficient) eat less
78 and spend less time eating (Young, Cai, & Dekkers, 2011) than pigs selected for high RFI
79 (HRFI; less efficient). In a previous study we showed that pigs from the more efficient line
80 were more immediately affected by weaning stress (growth check and more diarrhoea), but
81 managed to recover afterwards (Gilbert et al., 2019a). The present study was designed to
82 investigate the line differences in terms of behaviour, health and oxidative status, immune
83 and nutritional parameters of the pigs to understand the underlying mechanisms driving the
84 differences of robustness at weaning.

85

86 **Materials and methods**

87 The experiment was performed in INRAE experimental facilities in Saint-Gilles (France) in
88 compliance with the French directive on animal experimentation and care (2013-118) after
89 evaluation by the ethic committee in animal experimentation (C2EA – 07), and received an
90 authorization from the French Ministry of Research (project number 2016010512258334).

91

92 **Animals, housing, diets and experimental design**

93 Two Large White pig lines have been divergently selected for RFI (LRFI, pigs eating less
94 than predicted for their production level, and HRFI, pigs eating more than predicted) during 8
95 generations (Gilbert et al., 2007; Gilbert et al., 2017). Thirty six piglets from each line (total of
96 72 pigs, equal number of females and castrated males), all born during the same week, were
97 weaned at 28 days of age. The piglets were selected on the following criteria: maximisation
98 of the number of litters (9 sows for LRFI and 7 sows for HRFI), balanced number of castrated
99 males and females per litter, and average BW the day before the weaning (day-1) per line
100 representative of the average weaning BW in their contemporary line. Piglets did not have
101 access to creep feeding during lactation.

102 From the weaning day (day 0) until the end of the experiment (day 33), piglets were housed
103 in individual pens (80 x 60 cm, slatted floor) to measure individual feed intakes. Each pen
104 was equipped with a trough and a water nipple. Individual pens were separated by
105 transparent partitions (Plexiglass®) preventing physical contact with other pigs so limiting the
106 stress of individual housing but permitting visual contacts. The room temperature was
107 maintained at 28°C during the first week post-weaning and was then progressively
108 decreased to 24°C (1°C/day).

109 Pigs were fed a starter diet immediately after weaning, and a two days transition starting at
110 day 11 was applied between the starter and the weaning diets. Diets were provided by CCPA
111 (Janzé, France). Diets were based on wheat, barley, soybean meals and extruded soybean

112 seeds (see Control diets compositions in Gilbert et al., 2019a). They contained highly
113 digestive feedstuffs (cooked cereals, biscuits meals, whey, and potato proteins) to enhance
114 appetite and digestibility. The starter and weaning diets contained respectively 19.0 and 16.6
115 % of crude protein, 1.22 and 1.05 % of digestible lysine, and 10.44 and 9.70 MJ of net
116 energy. Titanium dioxide (0.4%) was included as an inert marker for digestibility calculations
117 in the two diets. The diets were offered as pellets. Pigs had free access to feed and water
118 throughout the experiment.

119

120 Observations, measurements, and sample collection

121 Individual feed intake (feed offered minus refusals) was recorded daily during the first three
122 weeks after weaning, and then weekly for the remainder of the experiment. Pigs were
123 weighed on days 0, 6, 12, 19, 26, and 33, and average daily gain was calculated for each
124 period. In addition, all piglets were individually weighed at birth for better understanding of
125 the general growth response of the animals. The faeces consistency was recorded at days 1,
126 2, 6, 12, and 19, using a three levels grid (normal, soft or diarrheic faeces). Rectal
127 temperature was measured on days 1, 5, 6 and 7 after weaning.

128 Behavioural observations of pigs were performed on 12 animals per line using 2-min
129 instantaneous scan sampling for 30 minutes per day (equivalent to 15 observations per day
130 for each pig), as previously described by Pastorelli et al. (2012). Behaviour of pigs was
131 observed during 4 periods : days 0 to 2, days 4 to 8, days 11 to 13 (feed transition), and days
132 18 to 21, excluding days when blood was sampled (days 6 and 18, details below). The scan
133 started 5 minutes after the feed distribution in the morning. Three postures (lying, sitting, and
134 standing) and five behaviours (inactive, eating, drinking, social behaviour like attempting to
135 sniff counterpart through the transparent side of the pen, and exploring) were recorded. The
136 traits observed during the 30 min scan sampling were expressed as a percentage of time
137 spent for each posture and activity. Then the percentage of time in each posture and activity
138 was calculated for each period of observation as the average of the percentages of the days
139 with observations.

140 Blood (5 mL into heparin and twice 5 mL into EDTA tubes) was collected at the jugular vein
141 the day before weaning (day-1), and at days 6 and 19, between 0800 and 1000 hours, before
142 feed distribution in the morning. Samples were kept on ice immediately after collection.

143 Plasma from one tube on EDTA was instantaneously used for blood formula analyses (white
144 and red cells counting). The two other tubes were centrifuged (2600 × g, 4°C, 10 min).

145 Plasma from the second EDTA tube was stored at -20°C for assays of glucose, non-
146 esterified fatty acid (NEFA), urea, insulin, IGF-1, immunoglobulins G and M, and haptoglobin.

147 Plasma from the heparin tube was stored at -80°C until analyses of the biological antioxidant
148 potential (BAP) and peroxide concentration (Reactive Oxygen Metabolites: dROM).

149 Approximately 10 g of faeces were collected at days 6 and 19 for volatile fatty acids (VFA)
150 and ammonia concentration assays. A phosphoric acid solution (0.5%, 1mL for 1g of faeces)
151 was immediately added to stop fermentation and samples were stored at -20°C. After faecal
152 collection for VFA and ammonia analyses on days 6 and 19, a plastic bag (stoma bag
153 Hollister, Paris, France; ref. 3778) was fixed on the animal anus using adhesive strip
154 (Tensoplast®, BSN Medical, Le Mans, France) to collect faeces on days 6 and 7, and 19 and
155 20 for digestibility analyses. The bag was removed after defecation and faeces were stored
156 at -20°C. After lyophilisation, samples of two consecutive days were pooled, grinded and
157 stored at +4°C.

158

159 Biological analyses

160 The total number of erythrocytes (red blood cells), leucocytes (white blood cells), and
161 neutrophil granulocytes was measured with a haematology automated cell counter calibrated
162 for pigs (MS9®; Melet Schloesing Laboratories, Osny, France). Kits were used to assay
163 plasma concentration of glucose (Glucose RTU; Biomérieux), NEFA (NEFA-HR(2), Wako
164 Chemicals GmbH), urea (UREA 981818, Thermo Fisher Scientific), and insulin (Insulin-CT,
165 IBA Molecular). All these compounds were analysed with Konélab 20XT automate (Thermo
166 Fisher Scientific, Courtaboeuf, France). Plasma IGF-1 concentration was determined after an
167 acid-ethanol extraction using the IRMA IGF-1 kit (Immunotech, Prague, Czech Republic).
168 Plasma haptoglobin was assayed by colorimetry (Tridelta PHASE Haptoglobin, Tridelta
169 Development). Immunoglobulins G (IgG) and M (IgM) were assayed by ELISA (Bethyl Elisa
170 quantitation; Interchim, Montluçon, France). The dROMs and BAP were assayed using a test
171 kit (Callegari, Parma, Italy; distributor Deltavit Laboratory, Janze, France) following the
172 method described by Sauerwein, Schmitz, & Hiss (2007). The concentration of
173 hydroperoxides generated by the peroxidation of lipids, proteins and nucleic acids (Alberti et
174 al., 1999) was measured with dROM. Results of the test were expressed in CARRU
175 (Carratelli Unit, 1 CARRU = 0.08 mg H₂O₂/100 mL of plasma). The BAP results from the
176 combined effects of antioxidants such as uric acid, ascorbic acid, proteins, alpha-tocopherol
177 and bilirubin (Benzie & Strain, 1996). Results are expressed in µmol.L of equivalent vitamin
178 C used as an iron-reducing reference agent. An Oxidative Stress Index (OSI) representing
179 the amount of oxidative products per anti-oxydant capacity was calculated as the ratio of
180 dROM to BAP (CARRU / µmol·L of Vit C-1; Sharma, Pasqualotto, Nelson, Thomas, &
181 Agarwal, 1999).

182 For the determination of VFA, frozen samples of faeces were first added (1 g/1 mL) with
183 0.4% (wt/vol) crotonic acid. After 2 h at 4°C, the samples were mixed and then twice
184 centrifuged at 16,500 × g for 10 min at 4°C. The VFA dissolved in the supernatant were then

185 determined by gas chromatography using 4-methylvaleric acid as internal standard (Jouany,
186 1982). Total concentration of VFA was expressed as a percentage of faecal DM.
187 Diets and faeces were analysed for DM (method 934.01), OM and minerals (method 942.05),
188 and crude proteins ($N \times 6.25$; method 990.03) using the AOAC (2000) methods. The
189 concentration of TiO₂ in diets and faeces was determined photometrically (Cobas Mira,
190 Horiba ABX, Montpellier, France) according to the method of Njaa (1961). The apparent total
191 tract digestibility of nutrients was calculated as previously described (Montagne et al., 2014).

192

193 Statistical analysis

194 All variables except faecal consistency scores were analysed with linear mixed models (lmer
195 function of lme4 package (Bates, Maechler, Ben Bolker, & Walker, 2015), lsmeans, car and
196 multcompView packages) of R software (R Core team 2015, version). Four pigs, two from
197 each line, were excluded from the experiment, three due to sanitary problem (acute
198 diarrhoea), and one due to persistent anorexia following weaning. The line, the time (period
199 or day), and the line by time interaction were used as fixed effects; the animal and the litter
200 were introduced as random factors. The body weight at weaning was used as a covariate for
201 performance traits. Pair-wise comparisons of least square means were performed using a
202 Tukey test. Chi² tests were used to test, at each day of observation, the effect of the line on
203 the number of pigs having normal, soft or diarrheic faeces. Differences with P-values lower
204 than 0.05 were considered as significant, and differences with P-values between 0.05 and
205 0.10 were reported as trends.

206

207 Results

208

209 Growth performance

210 The LRFI piglets were heavier than the HRFI piglets at weaning (9.08 ± 0.75 vs 7.96 ± 1.09
211 kg, $P < 0.005$, Table 1). A BW difference was already observed between lines at birth
212 (1.55 ± 0.23 and 1.33 ± 0.28 kg for LRFI and HRFI, respectively, $P < 0.01$). During the 33 days of
213 post-weaning period, BW adjusted to BW at weaning did not significantly differ between lines
214 ($P = 0.19$). The BW measured at days 12 and 19 were lower for LRFI pigs compared with
215 HRFI ($P = 0.001$). Other BW did not differ between lines ($P > 0.10$). Accordingly, a lower ADG
216 was observed for the LRFI line from days 0 to 11 (-40% compared with HRFI line, $P < 0.001$),
217 and lower average daily feed intake was observed in the same period (-34% , $P < 0.001$). The
218 difference in daily feed intake was observed until day 7 after weaning (Figure 1). For other
219 performances, there was no line difference in performance after day 12 (Table 1).

220

221 Digestion, fermentation and nutritional status

222 At the end of the first week after weaning (days 6 and 7), total tract apparent digestibility
223 coefficients of dietary components were significantly lower in pigs from the LRFI line
224 compared with HRFI pigs, from -6% for OM to -13% for N ($P < 0.001$; Table 2). Total tract
225 apparent digestibility coefficients increased with time and did not significantly differ between
226 lines on days 19 and 20, with an average of 82.2, 85.2, 60.7 and 79.6 % for dry matter,
227 organic matter, minerals, and N, respectively (see Table S1 for details).

228 The total concentration of VFA and ammonia did not differ between lines, and it increased
229 between days 6 and 19 ($P < 0.001$; Table 2). The ratio between total VFA and ammonia
230 tended to be lower for pigs of the LRFI line compared with HRFI pigs at the two times of
231 measurement ($P = 0.06$). The proportions of the different VFA differed with time ($P < 0.001$)
232 between the lines. In LRFI pigs, the proportion of acetate and branched chain fatty acids
233 were lower at day 19 than day 6, and those of propionate and butyrate were greater,
234 whereas in the HRFI pigs only the concentration of branched chain fatty acids significantly
235 changed between days 6 and 19 (see Table S2 for more details).

236 The time effect on plasma glucose concentration differed between the two lines ($P=0.02$). At
237 the day before weaning (day -1), LRFI pigs had greater plasma glucose concentration than
238 HRFI pigs (Table 2). Plasma glucose concentration was lower on day 6 compared with day -
239 1 for the two lines but did not differ between lines at days 6 and 19. The NEFA and urea
240 concentrations did not differ between lines the day before weaning, nor at day 19. By
241 contrast, greater concentrations were observed at day 6 in LRFI pigs (+167% for NEFA and
242 + 55% for urea compared to HRFI, $P < 0.001$). The IGF-1 concentrations were similar
243 between lines on days -1 and 19, but lower values were observed for the LRFI pigs
244 compared with HRFI pigs on day 6 ($P=0.001$). Plasma insulin concentration was affected
245 neither by the time nor by the line.

246

247 Fecal score, blood indicators of inflammation and oxidative status

248 Two episodes of diarrhoea were observed (Figure 2). During the first episode, from days 0 to
249 3 after weaning, the prevalence of pigs having diarrheic faeces was more important for the
250 HRFI line (12/34 pigs on day 3) than for the LRFI line (1/34 pigs) ($P<0.001$). Conversely,
251 during the second episode, from days 6 to 9, the prevalence of pigs with diarrhoea was
252 higher for the LRFI than for HRFI pigs (11 vs 2/34 pigs at day 7). The measurement of rectal
253 temperature did not evidence any fever episode (results not shown). Haptoglobin plasma
254 concentration increased from weaning to day 6 and decreased from day 6 to day 19
255 (Table 3), with a different time effect between lines ($P<0.001$): concentrations were greater
256 for the LRFI line at day 6 (+52% compared with HRFI).

257 The white blood cell counts differed with the two lines with a significant interaction between
258 time ($P=0.007$). In HRFI pigs, white blood cell counts increased from day -1 to 6, and was

259 unchanged at day 19, whereas it did not differ between day -1 and 6 and increased between
260 days 6 and 19 in the LRFI line. The red blood cell counts were overall greater in LRFI pigs
261 ($P=0.012$) and increased over time for both lines ($P= 0.023$). The concentrations of IgG did
262 not differ between lines just before weaning and on day 6, but lower concentrations were
263 measured in the plasma of LRFI pigs on day 19 (-35% of the concentrations of HRFI pigs,
264 $P<0.019$). Plasma concentration of IgM was lower the day before weaning in LRFI pigs
265 compared with HRFI pigs. It increased between days 0 and 6, and the difference between
266 lines was no more significant on days 6 and 19.

267 The concentrations of dROM changed similarly for the two lines ($P \geq 0.58$; Table 3) during
268 the experiment period ($P \leq 0.002$). The concentration of dROM increased from day -1 to day
269 6, and then remained unchanged between days 6 and 19. The anti-oxidative ability (BAP)
270 was similar between days -1 and 6 and tended to increase between day 6 and day 19.

271 Consequently, the ISO ratio was maximal at day 6 ($P<0.05$) and did not differ between the
272 lines.

273

274 Posture and activity traits

275 The time spent to postures and activities did not differ between lines the first week after
276 weaning (see Figure S1 for data). Pigs of the LRFI line tended to spend less time standing
277 from days 11-13 compared with HRFI (29% vs 42% of time, respectively; $P = 0.09$).

278 Inversely, the third week after weaning (days 18-21), pigs of the LRFI line spent more time
279 standing (48% vs 32% of the time for LRFI and HRFI respectively, $P<0.05$), whereas pigs of
280 the HRFI line stayed mainly lying (65% of the observation time vs 45% for HRFI and LRFI,
281 respectively, $P<0.05$, Figure S1a). The inactive time and the time spent on social behaviour
282 and exploration did not differ between lines at any period. The third week after weaning (days
283 18-21), LRFI pigs spent more time eating (38%) compared with HRFI pigs (24%, $P<0.01$,
284 Figure S1b).

285

286 Discussion

287 This study confirmed that the divergent selection for RFI during growth affected differently
288 the response of pigs to weaning. Pigs from the more efficient line (LRFI) were more affected
289 by weaning (growth check and more diarrhoea) during the first post-weaning week, and
290 managed to recover afterwards, as reported before (Gilbert et al., 2019a).

291

292 During the first week after weaning, the larger changes in BW observed for LRFI compared
293 with HRFI pigs suggested a greater sensitivity or a lower resistance to the weaning stress of
294 pigs selected for a greater feed efficiency. These differences might be partially explained by
295 a difference in voluntary feed intake. Actually, low and variable feed intake is a major cause

296 of post-weaning disorders and growth check (Pluske et al., 1997; Le Dividich & Sève, 2000).
297 In the present study the anorexia was probably greater because of the absence of creep
298 feeding during lactation. Both lines have low feed intake after weaning (less than 300 g/d per
299 pig), but the LRFI pigs ingested particularly little compared to HRFI pigs, despite a similar
300 time spent eating during the first week after the weaning. Note that behavioural observations
301 need to be interpreted with care because they result from punctual observations (scan) and
302 animals were housed in individual cases. In addition, this might have enhanced weaning
303 stress and limited stimulation by counterparts. The LRFI pigs spent probably more time on
304 feed investigation and rummage in the trough rather than feed ingestion. Differences in
305 feeding behaviours have been reported between these lines during the growing phase, with
306 HRFI pigs spending more time eating than LRFI pigs (Meunier-Salaün et al., 2011). Similar
307 differences were described in another experiment divergent selection in Yorkshire pigs
308 performed at the Iowa State University (Young et al., 2011).

309 Decrease in voluntary feed intake is a common animal response to a stressful event. We
310 hypothesize that LRFI pigs were more affected by the stress induced by weaning. Difference
311 in plasma cortisol in response to an ACTH injection was evidenced between HRFI and LRFI
312 pigs aged of 6 weeks (ie two weeks after weaning), indicating genetically driven line
313 differences in the adrenocortical axis reactivity. Pigs from both lines had similar basal cortisol
314 concentrations but the responses to ACTH of the LRFI pigs was greater and persisted longer
315 than that of HRFI pigs (Gilbert et al., 2019b) . Increased stimulation of the corticotropic axis is
316 a primary endocrine stress response pathway that has also consequence on metabolism,
317 inflammation and immunity (Mormède et al., 2011), and could also be partly responsible for
318 the greater response of LRFI pigs during the acute post-weaning phase.

319 The increase in plasma haptoglobin concentration between weaning and day 6 after weaning
320 was more marked in the LRFI pigs. Accordingly, as described by Dirkzwager, Veldman, &
321 Bikker (2005), the post-weaning anorexia leads to an inflammation of the intestinal mucosa
322 and a decrease of the digestive capacity that could be the causes of diarrhoea. As
323 inflammation is known to generate an oxidative stress on farm animals (Lykkesfeldt &
324 Svendsen, 2007), it could explain the increase of oxidative stress between days 0 and 6.
325 Such oxidative stress the first week after weaning is in accordance with Zhu et al. (2012) and
326 Buchet, Belloc, Leblanc-Maridor, & Merlot (2017). Even if the pro-oxidant molecules (dROM)
327 did not differ between lines at day 6, the increase from days 0 to 6 was consistently
328 numerically more marked for the LRFI pigs than the HRFI (+ 34% vs 25%).

329 These line differences can also be related to the greater incidence of diarrhoea in the LRFI
330 pigs observed from days 5 to 9 after weaning. Surprisingly, diarrhoeas were not associated
331 with an increase of white blood cells, contrasting with others studies (Le Huërou-Luron et al.,
332 2004). By contrast the number of white blood cells increased from day 0 to 6 in HRFI pigs.

333 That might be the consequence of the diarrheic episode observed in this line day 3 after the
334 the weaning. Diarrhoeas observed from days 5 to 9 after weaning in the LRFI pigs were not
335 associated with fever and might be the consequence of changes in the digestive capacity
336 and dysbiosis, rather than bacterial infection (Dirkzwager et al., 2005; Molist et al., 2014).
337 The lower total tract digestibility of nutrients in LRFI pigs compared with HRFI reinforced this
338 hypothesis of weak digestive capacities. The difference between the two lines was
339 particularly pronounced for nitrogen, with a difference of 12.9 units, which is twice the
340 difference measured for OM. Such a difference in total tract digestibility probably reflects a
341 difference in the true digestibility and so on availability of AA for proteins synthesis affecting
342 animal growth. Proteins that were not digested before the ileum were fermented in the distal
343 part of the gastro-intestinal tract with ammonia and branched chain VFA as the major end
344 products (Williams et al., 2001). The trend observed for greater concentration of branched
345 fatty acids and lower ratio between VFA and ammonia in the faeces of LRFI pigs at day 6 is
346 in accordance with the observed difference of nitrogen digestibility. A higher ratio between
347 VFA and ammonia is considered as favourable for the digestive health (Williams et al.,
348 2001). The lower ratio observed for the LRI line compared with HRFI could also be related to
349 the greater incidence of diarrhoea. Differences in the concentrations of fermentation end-
350 products in the faeces could be linked to differences in the microbiota composition observed
351 after weaning. Contrary to LRFI piglets whose fecal microbiota remained unchanged
352 between day 0 and day 6, the fecal microbiota in the HRFI line changed indicating probably a
353 faster adaptation to the new post-weaning conditions (Kubasova et al., 2018). After day 6 no
354 predominant genera relating to pathogenic strains were evidenced in these faeces. Because
355 changes in genera were often associated with post-weaning diarrhoea (Karasova et al.
356 2021), this reinforces the hypotheses of a nutritional rather than an infectious origin of
357 diarrhoea.

358 The lower feed intake and digestive capacity of LRFI might explain the nutritional status
359 difference of the two lines evidenced at day 6, and in turn the lower growth of LRFI pigs until
360 day 11. The greater plasma concentration of NEFA and urea measured in LRFI pigs may
361 result from a greater mobilization from body lipids and proteins, and a greater amino acid
362 catabolism for energy supply. Such differences between lines were also observed in Gilbert
363 et al. (2019). As the metabolism is considered to contribute to around 75% to the growth
364 check observed after weaning (Pastorelli, van Milgen, Lovatto, & Montagne, 2012), the
365 greater growth check in LRFI pigs was probably also greatly associated with greater
366 inflammation and diarrhoea, in association with changes in digestibility and in metabolic
367 status.

368

369 As indicated by the growth response to weaning, LRFI pigs had a greater sensitivity to
370 weaning but recovered afterwards and adapted to the post-weaning conditions. Three weeks
371 after weaning, the phenotypic differences between lines were limited: the lines had similar
372 growth performance, digestibility and VFA profiles, and were in a similar metabolic, oxidative
373 and health status. The genetic improvements of efficiency have been suggested to have
374 unfavourable consequences in the pig's ability to cope with stressors and to maintain their
375 health (Rauw, Kanis, Noordhuizen-Stassen, & Grommers, 1998; Doeschl-Wilson et al., 2009;
376 Prunier, Heinonen, & Quesnel, 2010). However, experimental challenges did not validate this
377 hypothesis in divergent RFI lines (Gilbert et al., 2017, Dunkelberger et al., 2015). When
378 exposed to various challenges, no advantage of one line or the other was previously
379 identified (response to inflammation and heat stress: Merlot, Gilbert, & Le Floc'h, 2016 ;
380 Renaudeau, Frances, Dubois, Gilbert, & Noblet, 2013; Campos et al., 2014 ; response to
381 weaning: Gilbert et al., 2019a), or there was an advantage to the LRFI pigs (Chatelet et al.,
382 2018 for a low hygiene challenge). In a similar and independent selection of divergent
383 selection on RFI on Yorkshire pigs (Cai, Casey, & Dekkers, 2018), lines were tested for their
384 ability to respond to an experimental infection by the PRRSV 1 to 3 weeks after weaning
385 (Dunkelberger et al., 2015) and with *Mycoplasma hyopneumoniae* and *Lawsonia*
386 *intracellularis* (Helm et al., 2018). The present study confirms that even if the short term
387 response to the weaning (day 6) differed between lines, the overall capability of pigs to cope
388 with the weaning disturbance, considered as a key component of robustness (Friggens et al.,
389 2017; Revilla et al., 2019) is similar in the two lines. This reinforces the idea that it is possible
390 to simultaneously improve productivity and robustness (Hermesch, Li, Doeschl-Wilson, &
391 Gilbert, 2015).

392

393 **Conclusion**

394 This study aimed at identifying the biological mechanisms underlying the temporal difference
395 of the response to weaning between lines. Selection on RFI has impacted the immediate
396 response but has globally not affected the robustness of pigs to the weaning challenge. Pigs
397 from the LRFI line, selected for greater feed efficiency during the growing period, were more
398 sensitive to the weaning stress with an important anorexia, growth check, and higher
399 frequency of diarrhoea at the end of the first week after weaning. There also seem to be
400 more resilient as they finally adapt to the new condition and recover to show similar
401 performance results as pigs of the HRFI line. Differences of the responses were related to
402 some indicators of digestion, to the nutritional, oxidative, inflammatory statuses and to some
403 fermentative indicators.

404

405 **Animal Welfare Statement**

406 The authors confirm that the ethical policies of the journal, as noted on the journal's author
407 guidelines page, have been adhered to and the appropriate ethical review committee
408 approval has been received. The authors confirm that they have followed EU standards for
409 the protection of animals used for scientific purposes.

410

411 **viii. References**

412 Alberti, A., Bolognini, L., Macciantelli, D., Caratelli, M. (1999). The radical cation of N,N-
413 diethyl-para-phenylendiamine: A possible indicator of oxidative stress in biological samples.
414 *Research on Chemical Intermediates*, 26, 253-267. doi: 10.1163/156856700X00769

415 AOAC. 2000. Official methods of analysis. 16th ed. The Association of Official Analytical
416 Chemists, Arlington, VA.

417 Bates, D., Maechler, M., Ben Bolker, B., & Walker, S. (2015). Fitting Linear Mixed-Effects
418 Models Using lme4. *Journal of Statistical Software*, 67, 1-48. doi:10.18637/jss.v067.i01

419 Benzie, I. F. F., & Strain, J. J. (1996). The ferric reducing ability of plasma (FRAP) as a
420 measure of "antioxidant power": The FRAP Assay. *Analytical Biochemistry*, 239, 70-76.
421 doi:10.1006/abio.1996.0292

422 Buchet, A., Belloc, C., Leblanc-Maridor, M., & Merlot, E. (2017). Effects of age and weaning
423 conditions on blood indicators of oxidative status in pigs. *PLoS ONE*, 12, e0178487.
424 doi:10.1371/journal.pone.0178487

425 Burrin, D., & Stoll, B. (2003). Intestinal nutrient requirements in weanling pigs. In: J.R.
426 Pluske, J. Le Dividich, M.W.A., Verstegen (Eds.), *Weaning the Pig: Concepts and*
427 *Consequences* (pp. 301-335). The Netherlands: Wageningen Academic Publishers.

428 Cai, W., Casey, D. S., & Dekkers, J. C. M. (2008). Selection response and genetic
429 parameters for residual feed intake in Yorkshire swine. *Journal of Animal Science*, 86, 287-
430 298. doi:10.2527/jas.2007-0396

431 Campos, P. H., Noblet, J., Jaguelin-Peyraud, Y., Gilbert, H., Mormède, P., de Oliveira
432 Donzele, R. F., Donzele, J. L., & Renaudeau, D. (2014). Thermoregulatory responses during
433 thermal acclimation in pigs divergently selected for residual feed intake. *International Journal*
434 *of Biometeorology*, 58, 1545-1557. doi:10.1007/s00484-013-0759-3

435 Chatelet, A., Merlot, E., Gilbert, H., Gondret, F., Friggens, N., & Le Floc'h, N. (2018). Impact
436 of hygiene of housing conditions on performance and health of two pig genetic lines
437 divergent for residual feed intake. *Animal*, 12, 350-358. doi:10.1017/S1751731117001379

438 Dirkzwager, A., Veldman, B., & Bikker, P. (2005). A nutritional approach for the prevention of
439 Post Weaning Syndrome in piglets. *Animal Research*, 54, 231–236.
440 doi:10.1051/animres:2005013

441 Doeschl-Wilson, A. B., Kyriazakis, I., Vincent, A., Rothschild, M. F., Thacker, E., & Galina-
442 Pantoja, L. (2009). Clinical and pathological responses of pigs from two genetically diverse
443 commercial lines to porcine reproductive and respiratory syndrome virus infection. *Journal of*
444 *Animal Science*, 87, 1638–1647. doi:10.2527/jas.2008-1447

445 Dunkelberger, J. R., Boddicker, N. J., Serao, N. V. L., Young, J. M., Rowland, R. R. R., &
446 Dekkers, J. C. M. (2015). Response of pigs divergently selected for residual feed intake to
447 experimental infection with the PRRS virus. *Livestock Science*, 177, 132-141.
448 doi:10.1016/j.livsci.2015.04.014

449 Friggens, N.C., Blanc, F., Berry, D.P., & Puillet, L. (2017). Review: Deciphering animal
450 robustness. A synthesis to facilitate its use in livestock breeding and management. *Animal*,
451 11, 2237-2251. doi:10.1017/S175173111700088X

452 Gilbert, H., Bidanel, J.- P., Gruand, J., Caritez, J. C., Billon, Y., Guillouet, P., Lagant, H.,
453 Noblet, J., & Sellier, P. (2007). Genetic parameters for residual feed intake in growing pigs,
454 with emphasis on genetic relationships with carcass and meat quality traits. *Journal of*
455 *Animal Science*, 85, 3182–3188. doi:10.2527/jas.2006-590

456 Gilbert, H., Billon, Y., Brossard, L., Faure, J., Gatellier, P., Gondret, F., Labussièrre, E.,
457 Lebreton, B., Lefaucheur, L., Le Floc'h, N., Louveau, I., Merlot, E., Meunier-Salaün, M.-C.,
458 Montagne, L., Mormède, P., Renaudeau, D., Riquet, J., Rogel-Gaillard, C., van Milgen, J.,
459 Vincent, A., & Noblet, J. (2017). Review: divergent selection for residual feed intake in the
460 growing pig. *Animal*, 11, 1427-1439. doi:10.1017/S175173111600286X

461 Gilbert, H., Terenina, E., Ruesche, J. , Gress, L., Billon, Y., Mormède, P., & Larzul, C.
462 (2018). Responses of pigs divergently selected for cortisol level or feed efficiency to a
463 challenge diet during growth. 11. World Congress on Genetics Applied to Livestock
464 Production (WCGALP), Feb 2018, Auckland, New Zealand. 1130 p. hal-01816156

465 Gilbert, H., Ruesche, J., Muller, N., Billon, Y., Begos, V., & Montagne, L. (2019). Responses
466 to weaning in two pig lines divergently selected for residual feed intake depending on diet.
467 *Journal of Animal Science*, 97, 43–54. doi:10.1093/jas/sky416

468 Helm E. T., Outhouse, A. C., Schwartz, K. J., Dekkers, J. C. M., Lonergan, S. M., Rauw, W.
469 M., & Gabler, N. K. (2018). Impact of *Mycoplasma hyopneumoniae* and *Lawsonia*
470 *intracellularis* on the performance of pigs divergently selected for feed efficiency. *Journal of*
471 *Animal Science*, 96, 462–472. doi:10.1093/jas/skx074

472 Hermesch, S., Li, L., Doeschl-Wilson, A., & Gilbert, H. (2015). Selection for productivity and
473 robustness traits in pigs. *Animal Production Science*, 55, 1437–1447. doi:[10.1071/an15275](https://doi.org/10.1071/an15275)

474 Jouany, J. P. (1982). Volatile fatty acid and alcohol determination in digestive contents,
475 silage juices, bacterial cultures and anaerobic fermentor contents. *Sciences des Aliments*, 2,
476 131-144.

477 Kubasova, T., Davidova-Gerzova, L., Darina Cejkova, D., Montagne, L., Le Floc'h, N., &
478 Rychlik, I. (2018). Effects of host genetics and environmental conditions on fecal microbiota
479 composition of pigs. *PLoS ONE*, 13, e0201901. doi:[10.1371/journal.pone.0201901](https://doi.org/10.1371/journal.pone.0201901)

480 Karasova, D., Crhanova, M., Babak, V., Jerabek M., Brzobohaty, L., Matesova, Z., & Rychlik,
481 I. (2021). Development of piglet gut microbiota at the time of weaning influences
482 development of postweaning diarrhea – A field study. *Research in Veterinary Science*, 135,
483 59-65, doi:[10.1016/j.rvsc.2020.12.022](https://doi.org/10.1016/j.rvsc.2020.12.022)

484 Lallès, J. P., & Guillou, D. (2015). Pig intestine, weaning and dietary interventions. In:
485 Niewold T. (Ed.), *Intestinal health. Key to maximise growth performance in livestock* (pp.139–
486 168). The Netherlands: Wageningen Academic Publishers. doi:[10.3920/978-90-8686-792-
487 9_6](https://doi.org/10.3920/978-90-8686-792-9_6)

488 Lallès, J. P., P. Bosi, H. Smidt, & Stokes, C. R. 2007. Weaning — A challenge to gut
489 physiologists. *Livestock Science*, 108, 82-93. doi:[10.1016/j.livsci.2007.01.091](https://doi.org/10.1016/j.livsci.2007.01.091)

490 Le Dividich, J., & Sève, B. (2000). Effects of underfeeding during the weaning period on
491 growth, metabolism, and hormonal adjustments in the piglet. *Domestic Animal
492 Endocrinology*, 19, 63–74. doi:[10.1016/S0739-7240\(00\)00067-9](https://doi.org/10.1016/S0739-7240(00)00067-9)

493 Le Huërou-Luron, I., Mroz, Z., Dekker, R., Koopmans, S. J., Zijlstra, R., Kogut, J., & Lelystad,
494 A. B. (2004). Effets d'un challenge infectieux, des conditions de logement et d'une
495 supplémentation en acidifiants sur les réponses physiologiques des porcelets au sevrage.
496 *Journées de la Recherche Porcine*, 36, 133–138.

497 Lykkesfeldt, J., & Svendsen, O. (2007). Oxidants and antioxidants in disease: Oxidative
498 stress in farm animals. *The Veterinary Journal*, 173, 502-511. doi:[10.1016/j.tvjl.2006.06.005](https://doi.org/10.1016/j.tvjl.2006.06.005)

499 Merlot, E., Gilbert, H., & Le Floc'h, N. (2016). Metabolic response to an inflammatory
500 challenge in pigs divergently selected for residual feed intake. *Journal of Animal Science*, 94,
501 563–573. doi:[10.2527/jas.2015-9445](https://doi.org/10.2527/jas.2015-9445).

502 Meunier-Salaün, M. C., Guérin, C., Billon, Y., Sellier, P., Noblet, J., & Gilbert, H. (2014).
503 Divergent selection for residual feed intake in group-housed growing pigs: characteristics of
504 physical and behavioural activity according to line and sex. *Animal*, 8, 1898–1906.
505 doi:[10.1017/S1751731114001839](https://doi.org/10.1017/S1751731114001839).

506 Molist, F., Van Oostrum, M., Pérez, J. F., Matéos, G. G., Nyachoti, C. M., & Van Der Aar, P.
507 J. (2014). Relevance of functional properties of dietary fibre in diets for weanling pigs. *Animal*
508 *Feed Science and Technology*, 189, 1–10. doi:10.1016/j.anifeedsci.2013.12.013.

509 Montagne, L., Boudry, G., Favier, C., Luron, I., Lallès, J.P., & Sève, B. (2007). Main intestinal
510 markers associated with the changes in the gut architecture and functions in piglets after
511 weaning. *British Journal of Nutrition*, 97, 45-57. doi:10.1017/S000711450720580X

512 Montagne, L., Loisel, F., Le Naou, T., Gondret, F., Gilbert H., & Le Gall M. (2014). Difference
513 in short-term response to a high fiber diet in pigs divergently selected on residual feed intake.
514 *Journal of Animal Science*, 92, 1512-1523. doi:10.2527/jas.2013-6623

515 Mormède, P., Foury, A., Barat, P., Corcuff, J. B., Terenina, E., Marissal-Arvy, N., & Moisan, M.
516 P. (2011). Molecular genetics of hypothalamic–pituitary–adrenal axis activity and function.
517 *Annals of the New-York Academy of Sciences*, 1220, 127–136. doi:10.1111/j.1749-
518 6632.2010.05902.x

519 Njaa, L. R. 1961. Determination of protein digestibility with titanium dioxide as indicator
520 substance. *Acta Agriculturae Scandinavica*, 11:227–241.
521 <https://doi.org/10.1080/00015126109434994>

522 Pastorelli, H., Le Floch, N., Merlot, E., Meunier-Salaün, M. C., van Milgen, J., & Montagne,
523 L. (2012). Sanitary housing conditions modify the performance and behavioural response of
524 weaned pigs to feed- and housing-related stressors. *Animal*, 6, 1811–1820.
525 doi:10.1017/S1751731112001231

526 Pastorelli, H., van Milgen, J., Lovatto, P., & Montagne, L. (2012). Meta-analysis of feed
527 intake and growth responses of growing pigs after a sanitary challenge. *Animal*, 6, 952–961.
528 doi:10.1017/S175173111100228X

529 Pluske, J. R., Hampson, D. J., & Williams, I. H. (1997). Factors influencing the structure and
530 function of the small intestine in the weaned pig: a review. *Livestock Production Science*, 51,
531 215–236. doi:10.1016/S0301-6226(97)00057-2

532 Prunier, A., Heinonen, M., & Quesnel, H. (2010). High physiological demands in intensively
533 raised pigs: impact on health and welfare. *Animal*, 4, 886-898.
534 doi:10.1017/S175173111000008X

535 Rauw, W. M., Kanis, E., Noordhuizen-Stassen, E. N., & Grommers, F. J. (1998). Undesirable
536 side effects of selection for high production efficiency in farm animals: a review. *Livestock*
537 *Production Science*, 56, 15–33. doi:10.1016/S0301-6226(98)00147-X

538 Renaudeau, D., Frances, G., Dubois, S., Gilbert, H., & Noblet, J. (2013). Effect of thermal
539 heat stress on energy utilization in two lines of pigs divergently selected for residual feed
540 intake. *Journal of Animal Science*, 91, 1162–1175. doi:10.2527/jas.2012-5689

541 Revilla, M., Friggens, N. C., Broudiscou, L. P., Lemonnier, G., Blanc, F., Ravon, L., Mercat,
542 M. J., Billon, Y., Rogel-Gaillard, C., & Le Floch, N. (2019). Towards the quantitative
543 characterisation of piglets' robustness to weaning: a modelling approach. *Animal*, 13, 2536-
544 2546. doi:10.1017/S1751731119000843

545 Sauerwein, H., Schmitz, S., & Hiss, S. (2005). The acute phase protein haptoglobin and its
546 relation to oxidative status in piglets undergoing weaning-induced stress. *Redox Report*, 10,
547 295-302. doi:10.1179/135100005X83725

548 Sauvart, D. & Martin, O. (2010). Robustness, rusticity, flexibility, placticity... the quality new
549 criteria of farm animals: systemic and biological definitions of the various concepts. *INRA*
550 *Productions Animales*, 23, 5-10. doi: 10.20870/productions-animales.2010.23.1.3280

551 Sharma, R. K., Pasqualotto, F. F., Nelson, D.R., Thomas, A.J., & Agarwal, A. (1999). The
552 reactive oxygen species—total antioxidant capacity score is a new measure of oxidative
553 stress to predict male infertility. *Human Reproduction*, 14, 2801–2807.
554 doi:10.1093/humrep/14.11.2801

555 Williams, B., Bosch, M. W., Awati, A., Konstantinov, S. R., Smidt, H., Akkermans, A.,
556 Verstegen M W. A., & Tamminga, S. (2005). In vitro assessment of gastrointestinal tract
557 (GIT) fermentation in pigs: Fermentable substrates and microbial activity. *Animal Research*,
558 54, 191–201. doi:10.1051/animres:2005011

559 Williams, B., Verstegen, M., & Tamminga, S. (2001). Fermentation in the large intestine of
560 single-stomached animals and its relationship to animal health. *Nutrition Research Reviews*,
561 14(2), 207-228. doi: <https://doi.org/10.1079/NRR200127>

562 Young, J. M., Cai, W., & Dekkers, J. C. M. (2011). Effect of selection for residual feed intake
563 on feeding behavior and daily feed intake patterns in Yorkshire swine. *Journal of Animal*
564 *Science*, 89, 639–647. doi:10.2527/jas.2010-2892

565 Zhu, L. H., Zhao, K. L., Chen, X. L., & Xu, J. X. (2012). Impact of weaning and an antioxidant
566 blend on intestinal barrier function and antioxidant status in pigs. *Journal of Animal Science*,
567 90, 2581–2589. doi:10.2527/jas.2012-4444

568

569

570 **ix. Tables**

571

572 Table 1. Performances (least-square means, n=34) after weaning (day 0) of pigs from two
573 lines divergently selected for residual feed intake (RFI)

	Line†			P-value‡			
	High RFI	Low RFI	SEM	Line	Time	Line×Time	BW ₀
BW (kg)							
Day 0 (weaning) §	8.77 ^g	8.32 ^g	0.24				
Day 6	9.41 ^{fg}	8.53 ^g	0.24				
Day 12	11.44 ^e	10.17 ^f	0.24	0.188	<0.001	0.119	<0.001
Day 19	13.97 ^c	12.80 ^d	0.23				
Day 26	17.28 ^b	16.15 ^b	0.24				
Day 33	21.53 ^a	20.63 ^a	0.24				
Average daily feed intake (g/d)							
Days 0 to 11	288 ^b	190 ^c	16				
Days 12 to 33	842 ^a	895 ^a	16	<0.001	<0.001	0.036	<0.001
Average daily gain (g/d)							
Days 0 to 11	239 ^b	139 ^c	13				
Days 12 to 33	498 ^a	483 ^a	13	<0.001	<0.001	<0.001	<0.001
Gain to Feed ratio							
Days 0 to 11	0.84 ^a	0.73 ^b	0.003				
Days 12 to 33	0.56 ^c	0.57 ^c	0.003	0.005	<0.001	0.007	0.419

574 † Two pig lines differing for their residual feed intake (RFI): less efficient High RFI vs more
575 efficient Low RFI576 ‡ Probability values for the effect of genetic line, time (days for BW or periods from 0 to 11
577 and 12 to 33 days for other criteria), interaction between line and time, and BW at weaning
578 (BW₀), introduced as a covariate in the statistical model579 §Average BW at weaning was 7.96±1.09 kg for High RFI pigs and 9.08±0.75 kg for Low RFI
580 pigs581 ^{a,g}Values within a trait with different superscripts differed significantly at P<0.05

582

583

584 Table 2. Apparent total tract digestibility coefficients, fermentation parameters and nutritional
 585 status (least-square means, n=34) after weaning (day 0) of pigs from two lines divergently
 586 selected for residual feed intake (RFI) †

	Line†			P-value‡		
	High RFI	Low RFI	SEM	Line	Time	Line×Time
Total tract digestibility of organic matter, %						
Day 6	81.3 ^b	75.0 ^c	0.83	<0.001	<0.001	<0.001
Day 19	85.4 ^a	84.9 ^a	0.76			
Total tract digestibility of nitrogen, %						
Day 6	72.1 ^b	59.2 ^c	1.80	<0.001	<0.001	<0.001
Day 19	80.3 ^a	78.9 ^a	1.72			
Total volatile fatty acids, µmol/g faeces						
Day 6	69.23 ^b	63.20 ^b	8.24	0.606	<0.001	0.858
Day 19	178.28 ^a	169.43 ^a	7.56			
Relative proportion of acetate/propionate/butyrate/ branched chain fatty acid, %§						
Day 6	61/20/9/7	63/18/9/8				
Day 19	60/22/11/5	59/22/11/5				
Ammonia, µmol/g faeces						
Day 6	18.84 ^b	24.22 ^b	3.49	0.276	<0.001	0.296
Day 19	46.64 ^a	45.02 ^a	3.20			
Glucose (mg/l)						
Day -1	1436.5 ^b	1654.4 ^a	36.8	<0.001	<0.001	0.021
Day 6	1234.7 ^c	1275.7 ^c	36.9			
Day 19	1242.3 ^c	1301.2 ^{bc}	37.7			
Non esterified fatty acid (µmol/l)						
Day -1	351.0 ^a	340.7 ^a	23.6	0.758	<0.001	<0.001
Day 6	129.6 ^b	346.9 ^a	23.8			
Day 19	38.5 ^b	39.2 ^b	24.5			
Urea (mg/l)						
Day -1	151.1 ^a	171.0 ^a	9.0	0.118	<0.001	0.002
Day 6	135.7 ^b	210.7 ^a	9.0			
Day 19	76.2 ^c	96.8 ^c	9.2			
Insulin (µU/l)						
Day -1	6.13	7.83	0.94	0.201	0.868	0.983
Day 6	6.73	8.12	0.94			
Day 19	6.68	8.26	0.97			

Insulin-like growth factor-1 (IGF-1) (ng/ml)

Day -1	152.2 ^b	161.3 ^b	7.9			
Day 6	107.7 ^c	57.3 ^d	8.0	0.418	<0.001	0.001
Day 19	200.6 ^a	177.4 ^{ab}	8.3			

587 † Two pig lines differing for their residual feed intake (less efficient High RFI vs more efficient

588 Low RFI)

589 ‡ Probability values for the effect of genetic lines, time (days of faeces or blood collection)

590 and interaction between line and time

591 § See supporting information Table S2 for more details

592 ^{a,d}Values within a criterion with different superscripts differed significantly at P<0.05

593

594 Table 3. Main indicators of health and oxidative status (least-square means, n=34) after
 595 weaning (day 0) of pigs from two lines divergently selected for residual feed intake (RFI) †

	Line†			P-value‡		
	High RFI	Low RFI	SEM	Line	Time	Line×Time
Haptoglobin (mg/ml)						
Day -1	0.36 ^c	0.24 ^c	0.14			
Day 6	1.53 ^b	2.33 ^a	0.14	0.561	<0.001	<0.001
Day 19	1.47 ^b	1.11 ^b	0.14			
White blood cell counts (10 ³ /mm ³)						
Day -1	14.47 ^d	15.57 ^{bcd}	0.95			
Day 6	17.81 ^{abc}	14.28 ^{cd}	0.95	0.414	0.001	0.007
Day 19	18.35 ^{ab}	19.01 ^a	0.97			
Red blood cell counts (10 ³ /mm ³)						
Day -1	6.91 ^b	7.23 ^{ab}	0.09			
Day 6	7.13 ^b	7.52 ^a	0.09	0.012	0.023	0.837
Day 19	7.18 ^{ab}	7.50 ^a	0.09			
Immunoglobulins G (g/l)						
Day -1	5.65 ^{ab}	4.58 ^{bcd}	0.32			
Day 6	5.47 ^{abc}	4.17 ^{cd}	0.32	0.019	0.151	0.062
Day 19	6.12 ^a	3.98 ^d	0.35			
Immunoglobulins M (g/l)						
Day -1	1.44 ^a	0.57 ^b	0.14			
Day 6	1.49 ^a	1.09 ^a	0.14	<0.001	0.751	0.054
Day 19	1.57 ^a	1.27 ^a	0.14			
Reactive Oxygen Metabolites (dROM), CARRU _s						
Day -1	469 ^b	428 ^b	23			
Day 6	590 ^a	574 ^a	23	0.217	<0.001	0.662
Day 19	585 ^a	582 ^a	24			
Biological Antioxidant Potential (BAP), μmol/L eq vit C						
Day -1	3467 ^{bc}	3436 ^{ca}	85.5			
Day 6	3494 ^{bc}	3599 ^{abc}	85.5	0.800	0.002	0.633
Day 19	3847 ^a	3813 ^{ab}	87.4			
Oxidative Stress Index¶						
Day -1	0.136 ^{bc}	0.127 ^c	0.006			
Day 6	0.159 ^a	0.151 ^{ab}	0.006	0.298	<0.001	0.584
Day 19	0.123 ^c	0.124 ^c	0.006			

596 † Two pig lines differing for their residual feed intake (less efficient High RFI vs more efficient
597 Low RFI)
598 ‡ Probability values for the effect of genetic lines, time (days of blood collection) and
599 interaction between lines and time
600 §CARRU = Carratelli Unit, 1 CARRU = 0.08 mg H₂O₂/100 ml of plasma; BAP = Biological
601 Antioxidant Potential
602 ¶Ratio between dROM and BAP
603 ^{a,d}Values within a criterion with different superscripts differed significantly at P<0.05
604

605

606 **x. Figures legends**

607

608 Figure 1. Average daily feed intake after weaning depending of the pig line (n=34).

609 Values are lsmeans and standard deviations (n=36).

610 The two pig lines were divergently selected on residual feed intake (RFI): less efficient High
611 RFI vs more efficient Low RFI.

612 #, *, ** and *** indicated trends or significant line differences at the indicated days with

613 $P < 0.10$, 0.05 , 0.001 and $P < 0.001$, respectively.

614

615 Figure 2. Number of piglets exhibiting diarrhoea after weaning depending of the pig line
616 (n=36).

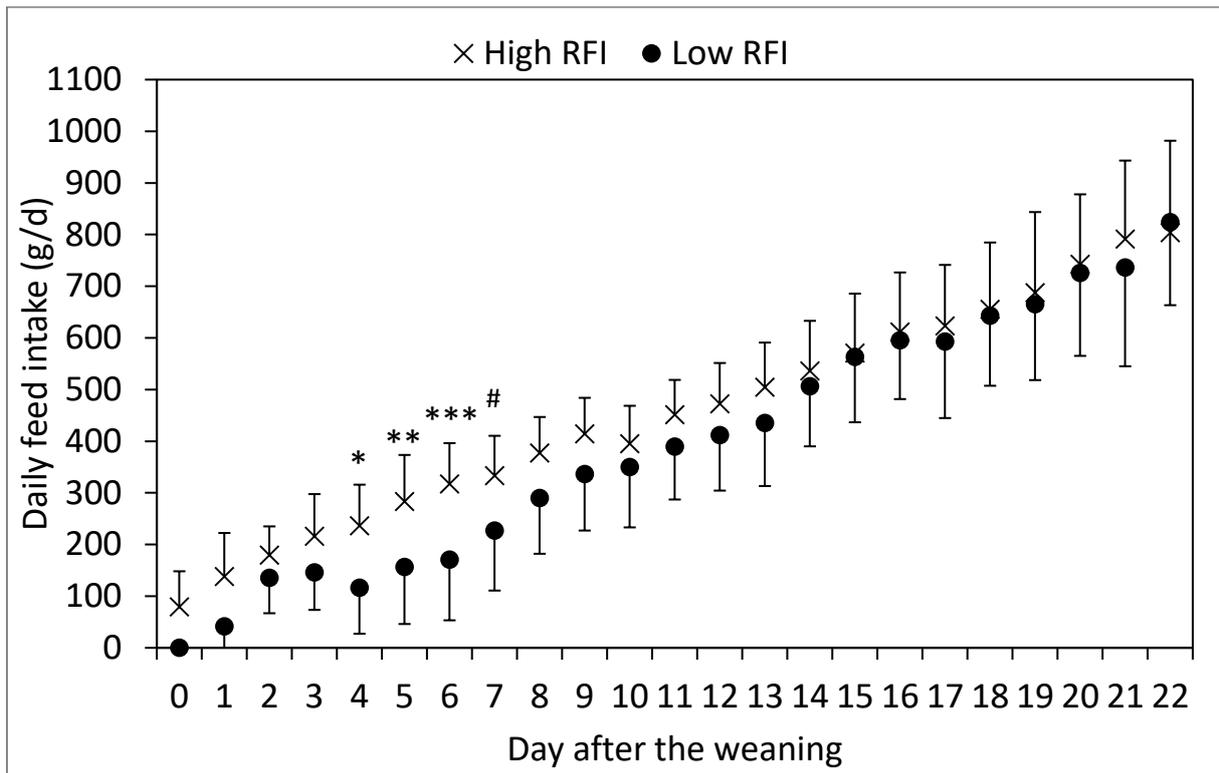
617 The two pig lines were divergently selected on residual feed intake (RFI): less efficient High
618 RFI vs more efficient Low RFI.

619 * and ** indicated significant line differences at the indicated days with $P < 0.05$ and $P < 0.01$,
620 respectively

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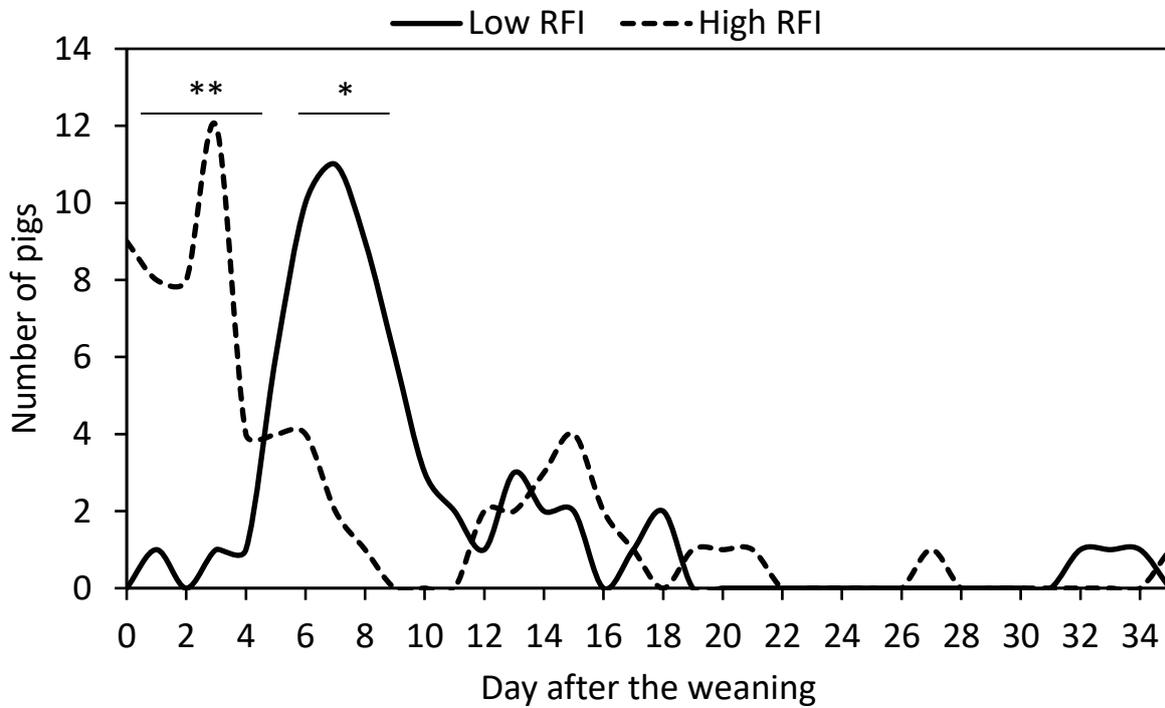
623 Fig 1



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626 Fig 2



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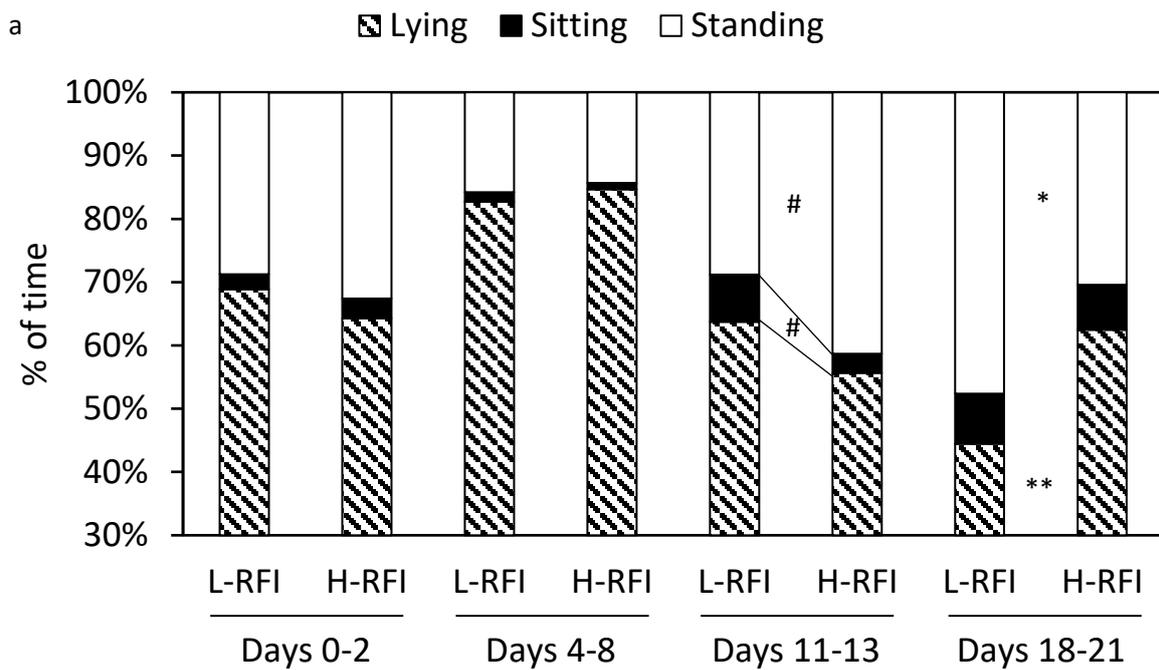
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630 Figure S1. Posture (a) and main activities (b) of pigs after weaning depending of the pig line
 631 (N=12).

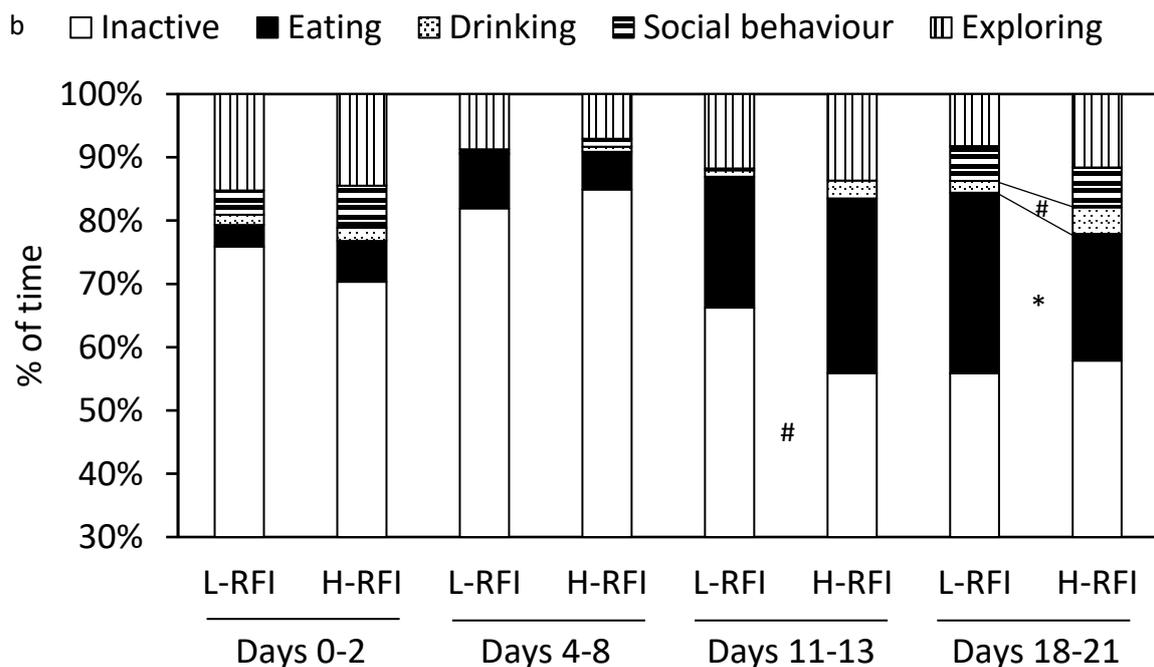
632 The two pig lines were divergently selected on residual feed intake (RFI): less efficient High
 633 H-RFI vs more efficient Low L-RFI.

634 Data are expressed as percentage of time spent in each posture and activity during each
 635 period of observation, based on 30 min scans per day.

636 #, *,** and *** indicated a trend or line difference at the indicated days with P<0.10, 0.05,
 637 0.001 and P<0.001, respectively.



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640 Table S1. Apparent total tract digestibility coefficients (least-square means, n=34) after
 641 weaning (day 0) of pigs from two lines divergently selected for residual feed intake (RFI) †

	Line†		SEM	P-value‡		
	High RFI	Low RFI		Line	Time	Line×Time
Dry matter						
Day 6	78.5 ^b	71.8 ^c	0.87			
Day 19	82.4 ^a	81.9 ^a	0.83	<0.001	<0.001	<0.001
Organic matter						
Day 6	81.3 ^b	75.0 ^c	0.83			
Day 19	85.4 ^a	84.9 ^a	0.76	<0.001	<0.001	<0.001
Minerals						
Day 6	40.0 ^b	29.1 ^c	1.75			
Day 19	60.6 ^a	60.7 ^a	1.628	<0.001	<0.001	<0.001
Nitrogen						
Day 6	72.1 ^b	59.2 ^c	1.80			
Day 19	80.3 ^a	78.9 ^a	1.72	<0.001	<0.001	<0.001

642 † Two pig lines differing for their residual feed intake (less efficient High RFI vs more efficient
 643 Low RFI)

644 ‡ Probability values for the effect of genetic lines, time (days of faeces collection) and
 645 interaction between lines and time

646 ^{a,c}Values within a criterion with different superscripts differed significantly at P<0.05

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650 Table S2. Dry matter, ammonia, total concentration and proportions of different volatile fatty
 651 acids (VFA, least-square means, n=34) after weaning (day 0) of faeces of pigs from the two
 652 lines divergently selected for residual feed intake (RFI) †

	Line†			P-value‡		
	High RFI	Low RFI	SEM	Line	Time	Line×Time
Total VFA, µmol/g faeces						
Day 6	69.23 ^b	63.20 ^b	8.24	0.606	<0.001	0.858
Day 19	178.28 ^a	169.43 ^a	7.56			
Acetate, %						
Day 6	61.37 ^{ab}	63.38 ^a	0.88	0.106	<0.001	0.057
Day 19	59.95 ^b	58.84 ^b	0.84			
Propionate, %						
Day 6	19.87 ^{bc}	18.08 ^c	0.55	0.021	<0.001	0.017
Day 19	21.55 ^{ab}	22.29 ^a	0.52			
Butyrate, %						
Day 6	9.28 ^{bc}	8.54 ^c	0.43	0.219	<0.001	0.228
Day 19	10.67 ^{ab}	10.93 ^a	0.41			
Branched chain fatty acid, %						
Day 6	6.76 ^a	7.59 ^a	0.33	0.075	<0.001	0.132
Day 19	5.29 ^b	5.29 ^b	0.31			
Ammonia, µmol/g faeces						
Day 6	18.84 ^b	24.22 ^b	3.49	0.276	<0.001	0.296
Day 19	46.64 ^a	45.02 ^a	3.20			
Ratio VFA by ammonia						
Day 6	4.32 ^{ab}	2.77 ^b	0.61	0.057	0.006	0.244
Day 19	5.22 ^a	4.93 ^a	0.52			

653 † Two pig lines differing for their residual feed intake (less efficient High RFI vs more efficient
 654 Low RFI)

655 ‡ Probability values for the effect of genetic lines, time (days of faeces collection) and
 656 interaction between lines and time

657 ^{a,c}Values within a criterion with different superscripts differed significantly at P<0.05

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