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1 **Intramuscular fat content in different muscles, locations, weights and**  
2 **genotype-sexes and its prediction in live pigs with computed tomography**

3 Maria Font-i-Furnols\*, Albert Brun, Marina Gispert

4 *IRTA-Food Industries, Finca Camps i Armet, 17121 Monells, Girona, Catalonia,*  
5 *Spain*

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8 **Short title** Intramuscular fat by muscle, weight & genotype-sex

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13 **Corresponding author:**

14 Maria Font-i-Furnols ([maria.font@irta.cat](mailto:maria.font@irta.cat)),

15 IRTA-Food Industries

16 Finca Camps i Armet

17 17121 Monells (Girona)

18 Spain

19 **Abstract**

20 Intramuscular (IMF) fat content depends on sex, genotype and diet, and varies  
21 with pig growth. The aim of the present work was to determine the evolution of  
22 IMF by genotype-sex, muscle, and muscle location, to determine relationships  
23 between IMF content of different muscles and to predict IMF in live pigs with  
24 computed tomography (CT). For this purpose, 155 pigs of seven combinations  
25 of genotype-sex were CT scanned and slaughtered at 70, 100 and 120 kg.  
26 From the carcasses fat thickness was measured at several locations along the  
27 midline. Loin samples from 3 anatomical positions (between the 8<sup>th</sup> and 9<sup>th</sup> last  
28 ribs, between the the 3<sup>rd</sup> and 4<sup>th</sup> last ribs, and between the 3<sup>rd</sup> and 4<sup>th</sup> *lumbar*  
29 *vertebrae*) and three ham muscles (*biceps femoris*, *semimembranous* and  
30 *gluteus medius*) were extracted, weighed and IMF was determined with near-  
31 infrared equipment. From CT images the distribution of volume by Hounsfield  
32 value (unit related with the density) was obtained for each muscle and  
33 anatomical location. Marbling was evaluated in the three loin locations. The  
34 effects of genotype-sex and live weight and their interaction were included in  
35 the statistical model. For prediction of IMF with CT images, partial least square  
36 regression was used. The results show differences in IMF content by genotype-  
37 sex and muscle. In general, the most cranial part of the loin presented higher  
38 IMF content, as well as the *biceps femoris* muscle of the ham. Depending on  
39 the genotype-sex, IMF content increased during all growth or increased until  
40 100 kg and then became constant. Correlation coefficients between IMF content  
41 by muscle/location were between 0.74 and 0.83 within loin locations and  
42 between 0.53 and 0.70 for ham muscles. Correlation coefficients between  
43 marbling and IMF content evaluated at the same location varied between 0.51

44 and 0.66. Prediction of IMF content from CT images is not accurate enough  
45 (residual predictive deviation statistical values lower than 1.3). Muscle weight  
46 increase with animal growth and allometric coefficients varied between 0.89 and  
47 0.97 for the muscles evaluated. The conclusions of the present work are that  
48 IMF content differs between and within muscle, during growth and by genotype-  
49 sex and that prediction of IMF in CT images of live pigs is not accurate.

50

### 51 **Keywords**

52 swine; marbling; loin; ham; growth

53

### 54 **Implications**

55 Intramuscular fat content is related to the eating quality of the meat and it is an  
56 important characteristic to improve meat quality. To know the pattern of muscle  
57 growth and deposition of intramuscular fat by genotype, sex and weight is  
58 important for breeding companies, producers and the meat industry to choose  
59 the best slaughter time to obtain the desired product regarding this  
60 characteristic. This work presents the intramuscular fat content and muscle  
61 weight by genotype-sex, muscle, muscle location and weight of the pig.

62

### 63 **Introduction**

64 Intramuscular fat (IMF) is an adipose tissue deposited within the lean tissue.  
65 Some previous works showed that its content depends on the breed or  
66 genotype (Plastow *et al.*, 2005, Tyra *et al.*, 2013), sex (Gispert *et al.*, 2010,

67 Jeong *et al.*, 2012, Škrlep *et al.*, 2012), diet (Huang *et al.*, 2008, Cordero *et al.*,  
68 2010, Lambe *et al.*, 2013) and age/weight (Bosch *et al.*, 2012, Tyra *et al.*,  
69 2013). However, some of these factors have not had a clear effect in other  
70 works; for instance, Jeong *et al.* (2012) did not find an effect of the diet and  
71 weight and Lambe *et al.* (2013) did not find an effect of weight. Additionally,  
72 there are differences in IMF content depending on the exact muscle (Lambe *et*  
73 *al.*, 2013, Tyra *et al.*, 2013) and within the same muscle (Faucitano *et al.*, 2004).  
74 Furthermore, some works show a relationship between IMF and meat  
75 palatability and eating quality and on consumers' acceptability (Brewer *et al.*,  
76 2001, Font-i-Furnols *et al.*, 2012) although this has not been confirmed in other  
77 works (Channon *et al.*, 2004, Moeller *et al.*, 2010). Because of the inconsistency  
78 in previous findings, it is important to increase knowledge about the growth of  
79 IMF tissue and its content depending on the muscle, genotype, sex and weight  
80 of the animals.

81 The knowledge about IMF content in live pigs is important mainly for breeding  
82 and nutritional purposes, and several technologies mainly based on ultrasound  
83 have been evaluated in live pigs and in carcasses with variable results (Newcom  
84 *et al.*, 2002, Lakshmanan *et al.*, 2012, Kvam and Kongsro, 2017). Computed  
85 tomography (CT) is a non-invasive technology based on X-rays used for breeding  
86 purposes. X-ray pass through the object and are attenuated in different degree  
87 depending on the density of the tissues they cross by and, this attenuation, is  
88 measured in Hounsfield units (HU). Lean has positive HU values from 0 to +140  
89 and fat has negative HU values from 0 to -149 approximately (Font-i-Furnols *et*  
90 *al.*, 2015), although the limits can change between works. As far as the authors  
91 know, only two works have studied the possibility of using CT to estimate IMF

92 content in live pigs, those from Kongsro and Gjerlaug-Enger (2013) that  
93 concluded that CT is not feasible for this purpose, and those from Lambe *et al.*  
94 (2013) that concluded that CT has a great potential to determine IMF in live pigs.  
95 Thus, further work is needed to evaluate the feasibility of CT to predict IMF in live  
96 pigs. The aim of the present work was to determine the patterns of IMF by  
97 genotype-sex, muscle and muscle location, to evaluate relationships between  
98 IMF content of different muscles and to predict IMF in live pigs with CT.

99

## 100 **Materials and Methods**

### 101 *Animals*

102 A total of 182 pigs were used that came from two experiments. Pigs were from  
103 seven combinations of genotype and sex: Pietrain × (Landrace × Large White)  
104 gilts (PL-F), Landrace × Large White gilts (LL-F), Duroc × (Landrace × Large  
105 White) gilts (DL-F), Pietrain × (Landrace × Duroc) gilts (PD-F), Pietrain ×  
106 (Landrace × Duroc) entire males (PD-M), Pietrain × (Landrace × Duroc)  
107 surgically castrated males (PD-C) and Pietrain × (Landrace × Duroc)  
108 immunocastrated males (PD-I) (2 vaccines, one at 12 weeks and the other one  
109 at 18 weeks of age) (Carabús *et al.*, 2015). There were no parental  
110 relationships within the breeds, except for PD-F, PD-M, PD-C and PD-I. Pigs  
111 were managed a standard way, fed a commercial diet *ad libitum*  
112 (Supplementary Material S1) and when they reach the desired live target body  
113 weight (TBW) they were CT scanned and, the same day after the scan, they  
114 were slaughtered. No withdrawal period was necessary because meat from pigs  
115 was not used for consumption. A total of 27 pigs (4–5 per genotype-sex, except

116 PD-I because at 30 kg immunocastration was not performed yet and they were  
117 the same as PD-M) were slaughtered at 30 kg (TBW), 31 pigs (4–5 per  
118 genotype-sex) were slaughtered at 70 kg TBW, 32 pigs (4–5 per genotype-sex)  
119 at 100 kg TBW and 92 pigs (11–16 per genotype-sex) at 120 kg (TBW) (Table  
120 1). Slaughter was performed at the experimental abattoir at IRTA Monells  
121 following standard commercial procedures and after stunning with CO<sub>2</sub>.

122

### 123 *Computed tomography scanning and image analysis*

124 Before slaughter live pigs of 70, 100 and 120 kg TBW were scanned with the  
125 General Electric HiSpeed Zx/I CT equipment placed at IRTA-CENTA (Monells).  
126 Scanning conditions were 140 kV, 145 mA, axials 1 s, 10 mm thick (Font-i-  
127 Furnols *et al.*, 2015). Before scanning pigs were anaesthetized by intramuscular  
128 injection of azaperone at 0.1 mg/kg and ketamine at 0.2 mg/kg, or intravenous  
129 injection of propofol at 0.2 mg/kg only at 100 and 120 kg TBW.

130 For the image analysis an in-house program of the software Matlab (Matlab  
131 Version 7.5.0.342 (R2007b) © The MathWorks, Inc.) was used. A region of  
132 interest (ROI) was selected for each of the studied muscles and/or locations  
133 (Figure 1). Three ROI's were studied for the *longissimus* muscle in three  
134 images, one between the 3<sup>rd</sup> and 4<sup>th</sup> *lumbar vertebrae*, one between the 3<sup>rd</sup> and  
135 4<sup>th</sup> last ribs and one between the 8<sup>th</sup> and 9<sup>th</sup> last ribs. Furthermore, from two  
136 images of the ham that allow to visualize the 3 muscles of interest, 3 different  
137 ROI's were performed; one image at the joint between the femur and the pubis  
138 bones for the muscle *gluteus medius* (GM), and one image in the middle of the  
139 femur for the muscles *semimembranosus* (SM) and *biceps femoris* (BF). For  
140 each ROI (n=923), the volume associated with each HU value was obtained by

141 means of the thickness of the image, the matrix size and the displayed field of  
142 view (Font i Furnols *et al.*, 2009) and its distribution was used for further  
143 analysis.

144

#### 145 *Carcass measures, muscle sampling and intramuscular fat analysis*

146 After slaughter fat thickness (plus skin) of the 70, 100 and 120 kg TBW pigs  
147 was measured perpendicularly to the skin with a ruler on the midline at three  
148 different levels of the loin (between the 3<sup>rd</sup> and 4<sup>th</sup> *lumbar vertebrae*—LD34LV,  
149 between the 3<sup>rd</sup> and 4<sup>th</sup> last ribs—LD34LR and between the 8<sup>th</sup> and 9<sup>th</sup> last  
150 ribs—LD89LR). Moreover, the minimum backfat and skin thickness over the GM  
151 muscle was also measured in the ham region.

152 Between 24 and 48 h after slaughter, the *longissimus* muscle and SM, GM and  
153 BF muscles of the ham were removed by trained butchers and weighed. Three  
154 samples of 4 cm thickness were obtained at different levels of the loin (LD34LV,  
155 LD34LR and LD89LR), minced, homogenized and a subsample of  
156 approximately 200 g was used for IMF analysis. IMF was also determined in the  
157 muscles SM, GM and BF from the ham using a subsample of approximately  
158 200 g obtained from mincing and homogenizing all the muscle. In the three loin  
159 samples, marbling was measured by two trained technicians according to the  
160 National Pork Producers Council (NPPC, 1999) scale (from 1=very low IMF to  
161 10=high IMF). Intramuscular fat of all of the muscles was determined using near  
162 infrared FoodScan equipment (Foss Analytical, Denmark). IMF for the 30 kg  
163 TBW pigs was not considered because the measures were not reliable.

164



165 *Statistical analysis*

166 Statistical analyses were performed with SAS software (version 9.3; SAS  
167 Institute Inc., Cary, NC, USA). The MEAN procedure was used to obtain mean  
168 and standard deviation values and the CORR procedure to determine  
169 Pearson's correlation between different variables. The analysis of variance was  
170 performed with the General Linear Model (GLM) procedure to determine the  
171 effect of the different factors on the IMF content. The model included genotype-  
172 sex, muscle and TBW as fixed effects. All of the double interactions and the  
173 triple interaction were added to the model. However, the triple interaction and  
174 the double interaction between muscle and TBW were removed because they  
175 were not significant (Supplementary Material S2). The GLM procedure was also  
176 used to determine the effect of the genotype-sex on the muscle weight. In this  
177 case, the model included the genotype-sex and the TBW as well as their  
178 interaction as fixed effects. To avoid the effect of differences in carcass weight  
179 within TBW, the difference between the average left carcass weight (within  
180 TBW) and the individual left carcass weight was included as a covariate  
181 (Supplementary Material S2). In all of the cases, significant differences between  
182 least square means were obtained after applying a Tukey test ( $P < 0.05$ ).

183 A prediction equation was obtained by partial least square regression (PLS) to  
184 predict the IMF content from the CT images of live pigs. Prediction was  
185 performed considering all of the muscles together and the muscles of the loin  
186 and the muscles of the ham separately. For this purpose, the PLS procedure  
187 was used and the volumes associated with the HU values of the ROIs were  
188 used as predictors. The smallest number of factors was selected to provide an  
189 error not significantly different ( $P > 0.1$ ) from the minimum error as suggested by

190 SAS. The root mean square error of prediction was calculated by cross-  
191 validation leave-one-out ( $RMSEP_{CV}$ ) following a modified version of the macro  
192 developed by Causeur *et al.* (2003) (see Supplementary Material S2).  
193 Additionally, to evaluate the predictive ability of the model, the residual  
194 predictive deviation statistics (RPD) was calculated as the standard deviation  
195 divided by the  $RMSEP_{CV}$ .

196

## 197 **Results**

198

### 199 *Intramuscular fat content by genotype-sex, live weight and muscle*

200 The averaged IMF content by muscle/location, genotype-sex and TBW is  
201 presented in Table 2. In the loin, it is possible to see that the mean IMF content  
202 at the LD34LR location varied between 1.02% (PL-F at 70 kg) and 2.22% (DL-F  
203 at 100 kg), at the LD34VL location the mean IMF content varied between 0.98%  
204 (PD-M at 70 kg) and 2.42% (DL-F at 100 kg), and at the LD89LR location the  
205 mean IMF content was higher in general and ranged between 1.34% (PD-F at  
206 100 kg) and 3.76% (DL-F at 100 kg). Regarding the ham muscles, SM  
207 presented mean IMF values between 1.11% (PD-M at 70 kg) and 2.60% (DL-F  
208 at 100 kg), BF between 1.31% (PD-M at 70 kg), and 2.73% (DL-F at 100 kg),  
209 and GM presented in general the lowest mean values of IMF, which varied  
210 between 1.10% (PD-M at 70 kg) and 2.27% (DL-F at 100 kg).

211 When the analysis of variance was performed for IMF content, a significant  
212 interaction was found between genotype-sex and muscle ( $P<0.0001$ ) and  
213 between genotype-sex and live TBW ( $P=0.0144$ ). The least squared means of

214 these interactions are shown in Table 3. It is possible to see that the highest  
215 IMF content was found in the LD89LR in DL-F, LL-F and PL-F, while in the  
216 other genotype-sexes this was not significantly different from those of BF and  
217 SM (and also GM in PD-M). Loin at location LD34LR presented the lowest IMF  
218 content in all of the genotype-sexes, although in some of them it was not  
219 significantly different from other locations. PD-C did not present significant  
220 differences in IMF content by TBW. However, for PD-F, PD-I, PD-M and PL-F,  
221 IMF content was increasing with the live weight of the animal. For DL-F pigs,  
222 IMF content was significantly higher in 100 and 120 kg than in 70 kg live weight  
223 and in LL-F, IMF was higher in 120 kg than in 100 kg live weight, being in  
224 between at 70 kg live weight. In fact, according to Table 2, were the raw mean  
225 and SD are presented, in LL-F, this occurred numerically with all of the muscles  
226 except BF and GM.

227

#### 228 *Relationship between intramuscular fat content and marbling by muscle*

229 Correlations of IMF content by muscle/location are presented in Table 4, with all  
230 of the correlations being significant ( $P < 0.0001$ ). The highest correlations were  
231 between IMF content at different locations of the loin (0.74 to 0.83). Correlations  
232 of IMF content between loin locations and ham muscles (0.53–0.70) were  
233 similar to those between ham muscles (0.63–0.67), being the lowest between  
234 LD34LR and SM (0.56) and between LD34LR and BF (0.53) and the highest  
235 between LD89LR and GM (0.70). The correlations by target body weight are  
236 presented in Supplementary Table S1 and Supplementary Table S2. These  
237 tables show that the correlations were lower in 70 kg live weight pigs than in

238 100 and 120 kg live weight pigs, maybe because the amount of IMF is lower at  
239 this weight for most of the genotype-sexes studied.

240 When correlations between marbling and IMF content are considered (Table 5)  
241 it is possible to see that the highest correlations were between marbling scores  
242 in the loin and IMF of the loin. The correlations between marbling and IMF in the  
243 same loin locations were between 0.51 and 0.66. As expected, the correlations  
244 were lower between marbling in the loin and IMF in the ham muscles.

245 Correlations between subcutaneous fat thickness at the three loin locations and  
246 at the ham with IMF content are also presented in Table 5. Although significant  
247 ( $P<0.0001$ ), the correlations are moderate, since except for those of IMF in GM  
248 muscle, all are lower than 0.50. Correlations of IMF in GM and the different  
249 subcutaneous fat thicknesses were between 0.52 and 0.54, being the highest  
250 between GM and subcutaneous fat thickness of the ham, which makes sense  
251 since it was measured over the GM muscle. These correlations for each target  
252 body weight are presented in Supplementary Table S3 to Supplementary Table  
253 S5. It is possible to see that in general, the highest correlations were found for  
254 100 kg live weight pigs.

255

#### 256 *Prediction of intramuscular fat content with computed tomography*

257 Prediction of IMF from CT images of live pigs had a RMSEP of 0.56–0.66,  
258 which according to a mean of 1.79–1.78% implies a coefficient of variation of  
259 31-37%. The prediction when all of the muscles are considered together  
260 (RPD=1.09,  $R^2=0.17$ ) is worse than those when only loin muscles are  
261 considered (RPD=1.28,  $R^2=0.42$ ). The worst prediction is when only ham

262 muscles are considered in the calculations (RPD= 1.03,  $R^2= 0.07$ ).  
263 Nevertheless, in all the cases RPD was much lower than 3, which is the  
264 recommended value for a suitable prediction model (Williams, 2001) (Table 6).  
265 In fact, the coefficient of determination was also very low, except for when only  
266 loin muscles were considered. The accuracy of the prediction was almost the  
267 same when only the Hounsfield values with variance importance for projection  
268 (VIP) higher than 0.8 were considered as predictors (results not shown) and,  
269 because of that, only the results of prediction using Hounsfield values between  
270 -50 and -120 are presented.

271

#### 272 *Muscles' weight by genotype-sex and target body weight*

273 Muscle growth with the live weight of the pig by genotype-sex is presented in  
274 Figure 2. In general, LL-F and DL-F genotypes-sex presented a lower weight of  
275 all the studied muscles than the other genotype-sex studied, especially at  
276 higher body weights. This difference can be seen also at early weights and it is  
277 maintained throughout growth. This is only a tendency in the muscle GM  
278 because no significant differences among genotypes-sexes can be seen in this  
279 muscle. The highest weights of the muscles were usually by the PD-F  
280 genotype-sex, although in most of the muscles/locations, not significantly  
281 different than those from the other genotype lines with Pietrain. The average  
282 allometric coefficient of the total muscle weight with respect to the live weight of  
283 the pig was 0.97 for the loin, 0.89 for SM, 0.95 for BF and 0.92 for GM, although  
284 there were some differences between genotypes-sex, with DL-F, LL-F and PL-F  
285 having the lowest allometric coefficients (results not shown).

287 **Discussion**

288 Intramuscular fat content varied between muscles (Table 3). Furthermore,  
289 within some muscles, such as *longissimus dorsi*, IMF also varied depending on  
290 its anatomical position. In this work, we confirmed what was presented by  
291 Faucitano *et al.* (2004) for *longissimus* muscle. The IMF content was higher  
292 between the 8<sup>th</sup> and 9<sup>th</sup> last ribs, then it decreased at the level of the 3<sup>rd</sup>–4<sup>th</sup> last  
293 ribs and then it increased again, between the 3<sup>rd</sup> and 4<sup>th</sup> *lumbar vertebrae*.  
294 Thus, IMF content was much higher at the most cranial part of the loin (8<sup>th</sup>–9<sup>th</sup>  
295 last ribs). Regarding IMF in three ham muscles, there were also some  
296 differences and, in general, BF presented the highest IMF values. Lambe *et al.*  
297 (2013) found higher levels of IMF in SM muscle compared with LT muscle at the  
298 last rib level (1.79 vs 1.18%, respectively), which is in agreement with the  
299 present results, but Tyra *et al.* (2013) found the opposite, high IMF levels in LT  
300 (at the last rib level) compared to SM (1.95 vs. 1.76%).

301 Intramuscular fat content for all of the muscles was higher in PD-C compared  
302 with PD-F, PD-I and PD-M, indicating a higher IMF content for castrated pigs, in  
303 accordance with Faucitano *et al.* (2004), Gispert *et al.* (2010) and Jeong *et al.*  
304 (2012), among others. Furthermore, IMF content depended on the genotype,  
305 with crosses with Duroc higher (DL-F) than those from Landrace-Large White  
306 crosses (LL-F) and crosses with Pietrain (PL-F). This is in accordance with the  
307 results found by Plastow *et al.* (2005) and Gil *et al.* (2008) studying pure breeds.  
308 Pietrain are animals with high muscularity (Plastow *et al.*, 2005, Gispert *et al.*,  
309 2010) and from very lean breed. This high muscularity is related with a low IMF  
310 content (Hocquette *et al.*, 2010).

311 For most of the genotype-sex groups (PD-F, PD-I, PD-M and PL-F) the IMF  
312 increased with the growth of the pig, indicating that the highest IMF content is  
313 reached at 120 kg, with a content significantly higher than that found at 70 kg  
314 (Table 3). These results are in agreement with those found by Bosch *et al.*  
315 (2012) when pigs of 95, 113 and 105 kg were studied. A tendency can also be  
316 seen for PD-C but not significant differences between weights and, for DL-F, at  
317 100 kg the IMF stops increasing. Bosch *et al.* (2012) did not find significant  
318 differences in the IMF content of (Yorkshire × Landrace) × Duroc gilts and  
319 barrows at 110, 125 and 138 kg. Thus, for these genotype-sexes, the early  
320 development of IMF would avoid the necessity of increasing the weight of the  
321 pig to increase its IMF content in all of the muscles. However, there was an  
322 opposite finding for LL-F, and the difference in IMF is very important ( $P<0.05$ )  
323 between 100 and 120 kg. Lambe *et al.* (2013) for Pietrain × (Landrace × Large  
324 White) intact male pigs fed different diets found no important differences in IMF  
325 content in pigs after 60 kg of weight, since the average IMF content was 1.14,  
326 1.12 and 1.18% for pigs of 60, 85 and 115 kg, respectively. Thus, in general, in  
327 the present study, the higher the weight (up to 120 kg) the higher the IMF  
328 content with some exceptions. In this sense, it is important to know the  
329 genotype-sex of the pigs to optimize the IMF content of the final product and to  
330 decide the optimal slaughter moment to obtain the desired product.

331 Since the IMF content is related to the tenderness and juiciness of the meat  
332 (Enfält *et al.*, 1997, Heyer and Lebret, 2007, Font-i-Furnols *et al.*, 2012), it is  
333 important to know the differences in this content between genotypes-sex and  
334 within muscles, because this can help in understanding the eating  
335 characteristics of the meat used for consumption. Even though there are

336 variations between and within muscles in the IMF content, correlations of IMF  
337 content within the same muscle (*longissimus*) are high (0.74–0.83) (Table 4).  
338 Correlations between muscles of the same animal were always significant and  
339 varied between 0.53 and 0.70 (Table 4). These correlations were higher when  
340 only 100 kg live weight pigs were considered and lower when only 70 kg live  
341 weight pigs were taken into account (Supplementary Table S1). This result may  
342 indicate that the knowledge of the IMF content of one muscle can be related to  
343 the other muscles, since when it increases, it increases for all of the muscles;  
344 nevertheless, the relationship is not perfect and it would be difficult to estimate  
345 the IMF content of one muscle from those of other muscles.

346 The correlation between marbling evaluated according to the NPPC scale and  
347 IMF content is higher for the *longissimus* than for the muscles of the ham. This  
348 is probably because the NPPC scale was measured in the *longissimus* muscle  
349 and, consequently, the similarities are higher. These correlations were always  
350 positive and significant, indicating the higher the IMF, the higher the marbling.  
351 Nevertheless, the correlation is not perfect, varying from 0.51 to 0.66 in the loin  
352 and between 0.33 and 0.50 in the ham muscles. These correlations are lower  
353 than those reported by Font-i-Furnols *et al.* (2012), which was 0.89, even in 100  
354 kg live weight animals that presented the highest correlations (Supplementary  
355 Tables S3 to S5).

356 The weight of the *longissimus* muscle was the highest followed by BF, SM and  
357 GM (Figure 2), but logically all of the muscles increase in weight when an  
358 animal grows. Except for the GM muscle, differences in weight for the same  
359 muscle between genotype-sex can be seen at 30 kg TBW and this became  
360 more important at 120 kg, where differences are greater. Crosses that do not



361 include Pietrain (LL-F and LL-F) presented in general a lower weight of the  
362 muscles, especially at higher TBW. This is in agreement with the work of Fisher  
363 *et al.* (2003) and Carabús *et al.* (2014) where the ham muscles of several  
364 genetic types were studied. This makes sense since Pietrain is a highly  
365 conformed breed, whose carcasses have higher ham weight, ham lean content  
366 and loin lean content, although not loin weight (Gispert *et al.*, 2007). Thus, the  
367 most adequate genotype-sex depends on the desired final product. Fisher *et al.*  
368 (2003) predicted ham muscle weights for carcasses of approximately 25 kg and  
369 75 kg. For 25 kg carcasses, which would represent 31 kg live weight, the  
370 average weight of the muscles SM (367 g) and GM (239 g) are similar to those  
371 found in the present project (367 g and 239 g, respectively). In 75 kg carcasses,  
372 which would represent approximately 93 kg live weight, the average weight of  
373 the muscles SM (833 g) and GM (617 g) are similar to those of 70 kg of the  
374 present work (844 g and 616 g, respectively). Thus, it seems that the growth of  
375 these muscles, at higher weights, is higher in the present work, probably due to  
376 differences in the genotypes used. Moreover, the allometric coefficients for ham  
377 (0.89-0.95) and loin (0.97) muscle weight with respect to the live weight of the  
378 pig were close to one in agreement with previous works that reported allometric  
379 coefficients between 0.87 and 1.13 for ham muscle and 1.01 and 1.03 for loin  
380 muscle (Carabús *et al.*, 2014, Carabús *et al.*, 2017).

381 The estimation of IMF content in live pigs is very important to optimize the  
382 slaughter moment of the pig to yield the final desired product. However, this  
383 prediction in live pigs by means of CT, in the conditions of this experiment, was  
384 not accurate enough and had an unacceptable error (Table 6). This result is in  
385 accordance with those reported by Kongsro and Gjerlaug-Enger (2013) who

386 concluded that CT is not a feasible method for prediction of IMF from live pigs.  
387 This is probably due to the respiratory movement of the pig and to the size of  
388 the voxel that is probably too big because the displayed field of view has to  
389 cover all of the body of the pig and, since the IMF is small, it might have many  
390 partial volume effect that makes it difficult to identify the IMF. These two  
391 reasons can explain why, although still not good, the prediction of IMF in loins is  
392 somewhat better (Font-i-Furnols *et al.*, 2013), since there is no breath  
393 movement and the displayed field of view can be adjusted to the loin area.  
394 Furthermore, the prediction of IMF is better when only loin muscles images are  
395 considered in the analysis compared with those of ham muscles. This is  
396 probably because for loin muscles, the sample analysed for IMF is almost the  
397 same as those analysed in CT images. However, for ham muscles, the whole  
398 muscle was minced and the IMF determined, while only one image of the  
399 muscle was analysed to predict IMF content. However, Lambe *et al.* (2013)  
400 predicted IMF from CT images using the average muscle density as a predictor.  
401 The error of prediction was not provided, but the correlation between this  
402 density and the chemical IMF was found to be around  $-0.44$  for 60 kg pigs,  
403  $-0.63$  for 85 kg pigs and  $-0.70$  for 115 kg pigs. Thus, these correlations indicate  
404 that CT can predict IMF, but the error is probably quite high, especially in pigs of  
405 lower weight. In the present project the correlation between averaged  
406 Hounsfield value of the ROI of each muscle and IMF content was 0.12 (0.16 at  
407 70 kg, 0.24 at 100 kg and 0.15 at 120 kg) and, because of that, this approach  
408 for IMF prediction has not been considered.

409 In the conditions of the present experiment, it can be concluded that the IMF  
410 content differs between and within muscle, during growth and by genotype-sex.

411 To optimize the final product for IMF content and amount of muscle it is  
412 important to know the growth of these tissues with the growth of the pig.  
413 Nevertheless, this is difficult to obtain from live pigs by means of computed  
414 tomography since the prediction of this parameter is not accurate enough.

415

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424

#### 425 **Declaration of interest**

426 The authors have no conflicts of interest.

427

#### 428 **Ethics statement**

429 The IRTA's ethical committee approved the protocol.

430

#### 431 **Software and data repository resources**

432 The data are not deposited in an official repository.

433

434

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544 **Table 1** *Number of pigs scanned, slaughtered and sampled (muscle weight and*  
545 *intramuscular fat-IMF) by genotype-sex and target body weight (TBW), total*  
546 *number of muscles evaluated by genotype-sex and mean live weight and warm*  
547 *carcass weight by TBW.*

Genotype-sex <sup>3</sup>	TBW				Total <sup>2</sup>
	30 kg <sup>1</sup>	70 kg	100 kg	120 kg	
PD-C	4	4	4	12	120
PD-F	4	4	4	12	120
PD-I <sup>4</sup>	-	4	4	12	120
PD-M	4	4	5	11	120
DL-F	5	5	5	16	156
LL-F	5	5	5	15	150
PL-F	5	5	5	14	144
TBW (kg) <sup>5</sup>	30.6±1.83	69.15±8.10	100.8±2.87	120.9±6.5	
Carcass weight (kg) <sup>5</sup>	23.4±1.60	56.5±2.23	81.8±2.52	98.4±2.85	

<sup>1</sup>: At 30 kg only the weight of the muscles was measured.

<sup>2</sup>: Total numbers of samples evaluated for IMF considering that for each genotype-sex and TBW, 6 different muscles/locations were analysed.

<sup>3</sup>: PD-C: Pietrain × (Landrace × Duroc) surgically castrated males; PD-F: Pietrain × (Landrace × Duroc) gilts; PD-I: Pietrain × (Landrace × Duroc) immunocastrated males; PD-M: Pietrain × (Landrace × Duroc) entire males; DL-F: Duroc × (Landrace × Large White) gilts; LL-F: Landrace × Large White gilts; PL-F: Pietrain × (Landrace × Large White) gilts.

<sup>4</sup>: At 30 kg, the immunocastrated pigs were entire male pigs.

<sup>5</sup>: Mean ± SD

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551 **Table 2** Mean and standard deviation (in italics) of the intramuscular fat content  
 552 (%) by pig genotype-sex (G-S), muscle/location<sup>1</sup> and target body weight (TBW).

G-S <sup>2</sup>	TBW	LD89LR	LD34LR	LD34VL	SM	BF	GM
	70	1.95±0.74	1.35±0.27	1.44±0.18	1.83±0.54	1.98±0.36	1.51±0.16
PD-C	100	1.84±0.52	1.53±0.50	1.75±0.52	2.08±0.46	2.30±0.50	1.70±0.52
	120	2.31±0.81	1.40±0.42	1.58±0.43	1.69±0.54	2.14±0.39	1.68±0.35
	70	1.58±0.65	1.03±0.12	1.15±0.06	1.35±0.19	1.39±0.33	1.22±0.09
PD-F	100	1.34±0.41	1.04±0.34	1.12±0.32	1.78±0.50	1.93±0.51	1.23±0.15
	120	1.79±0.61	1.10±0.26	1.22±0.27	1.75±0.35	2.07±0.51	1.43±0.25
	70	1.43±0.17	1.03±0.20	1.08±0.16	1.20±0.15	1.36±0.25	1.13±0.24
PD-I	100	1.75±0.48	1.11±0.23	1.28±0.36	1.33±0.24	1.76±0.39	1.28±0.32
	120	1.86±0.76	1.22±0.46	1.42±0.56	1.61±0.28	1.95±0.41	1.53±0.34
	70	1.36±0.67	1.09±0.47	0.98±0.34	1.11±0.32	1.31±0.35	1.10±0.35
PD-M	100	1.53±0.49	1.04±0.34	1.10±0.28	1.43±0.32	1.47±0.21	1.33±0.32
	120	1.68±0.50	1.08±0.26	1.19±0.22	1.62±0.39	1.91±0.48	1.43±0.41
	70	3.27±0.13	2.20±0.13	1.88±0.36	1.30±0.36	1.72±0.51	1.34±0.18
DL-F	100	3.76±1.20	2.22±0.97	2.42±0.95	2.60±0.51	2.73±0.57	2.27±0.96
	120	3.41±1.01	2.09±0.74	2.38±0.72	2.37±0.75	2.49±0.73	2.20±0.55
	70	2.49±0.82	1.59±0.56	1.70±0.12	1.91±0.72	1.78±0.39	1.41±0.41
LL-F	100	2.46±0.51	1.48±0.65	1.27±0.61	1.55±0.47	2.00±0.13	1.57±0.92
	120	2.66±0.64	1.54±0.42	1.77±0.48	2.14±0.40	2.14±0.62	1.95±0.43
	70	2.12±0.54	1.02±0.20	1.47±0.25	1.27±0.35	1.53±0.77	1.17±0.42
PL-F	100	2.19±0.98	1.35±0.30	1.51±0.47	1.72±0.38	1.92±0.81	1.72±0.29
	120	2.91±0.76	1.34±0.64	1.60±0.42	1.76±0.51	1.87±0.61	1.84±0.49

553 <sup>1</sup>LD34LR: between the 3<sup>rd</sup> and 4<sup>th</sup> last ribs; LD34VL: between the 3<sup>rd</sup> and 4<sup>th</sup> lumbar vertebrae;

554 LD89LR: between the 8<sup>th</sup> and 9<sup>th</sup> last ribs; BF: *Biceps femoris*; GM: *Gluteus medius*; SM:

555 *Semimembranosus*

556 <sup>2</sup> PD-C: Pietrain × (Landrace × Duroc) surgically castrated males; PD-F: Pietrain × (Landrace ×  
557 Duroc) gilts; PD-I: Pietrain × (Landrace × Duroc) immunocastrated males; PD-M: Pietrain ×  
558 (Landrace × Duroc) entire males; DL-F: Duroc × (Landrace × Large White) gilts; LL-F: Landrace  
559 × Large White gilts; PL-F: Pietrain × (Landrace × Large White) gilts.

560 **Table 3** Least square means of intramuscular content (%) for the interaction  
 561 between muscle or target body weight (TBW) with pig genotype-sex (RMSE =  
 562 0.52%)+.

	Genotype-sex						
	PD-C	PD-F	PD-I	PD-M	DL-F	LL-F	PL-F
Muscle (genotype-sex x muscle, $P < 0.0001$ )							
LD89LR	2.14 <sup>a</sup>	1.60 <sup>ab</sup>	1.68 <sup>ab</sup>	1.52 <sup>a</sup>	3.40 <sup>a</sup>	2.51 <sup>a</sup>	2.52 <sup>a</sup>
LD34LR	1.41 <sup>b</sup>	1.02 <sup>c</sup>	1.09 <sup>c</sup>	1.02 <sup>c</sup>	2.08 <sup>b</sup>	1.46 <sup>c</sup>	1.20 <sup>c</sup>
LD34VL	1.58 <sup>b</sup>	1.13 <sup>c</sup>	1.25 <sup>c</sup>	1.06 <sup>bc</sup>	2.24 <sup>b</sup>	1.59 <sup>bc</sup>	1.48 <sup>bc</sup>
SM	1.79 <sup>ab</sup>	1.62 <sup>ab</sup>	1.39 <sup>abc</sup>	1.41 <sup>a</sup>	2.16 <sup>b</sup>	1.92 <sup>b</sup>	1.58 <sup>bc</sup>
BF	2.13 <sup>a</sup>	1.85 <sup>a</sup>	1.72 <sup>a</sup>	1.62 <sup>a</sup>	2.34 <sup>b</sup>	1.96 <sup>b</sup>	1.73 <sup>b</sup>
GM	1.64 <sup>b</sup>	1.29 <sup>bc</sup>	1.32 <sup>bc</sup>	1.28 <sup>abc</sup>	1.99 <sup>b</sup>	1.69 <sup>bc</sup>	1.60 <sup>bc</sup>
TBW (genotype-sex x TBW, $P = 0.0144$ )							
70 kg	1.68	1.29 <sup>b</sup>	1.20 <sup>b</sup>	1.16 <sup>b</sup>	1.95 <sup>b</sup>	1.81 <sup>ab</sup>	1.43 <sup>b</sup>
100 kg	1.86	1.41 <sup>ab</sup>	1.42 <sup>ab</sup>	1.32 <sup>ab</sup>	2.67 <sup>a</sup>	1.72 <sup>b</sup>	1.74 <sup>ab</sup>
120 kg	1.80	1.56 <sup>a</sup>	1.60 <sup>a</sup>	1.48 <sup>a</sup>	2.49 <sup>a</sup>	2.03 <sup>a</sup>	1.89 <sup>a</sup>

+Different superscripts within genotype-sex indicate significant differences ( $P < 0.05$ ) between muscle or between TBW

LD34LR: between the 3<sup>rd</sup> and 4<sup>th</sup> last ribs; LD34VL: between the 3<sup>rd</sup> and 4<sup>th</sup> lumbar vertebrae; LD89LR: between the 8<sup>th</sup> and 9<sup>th</sup> last ribs; BF: *Biceps femoris*; GM: *Gluteus medius*; SM: *Semimembranosus*

PL-F: Pietrain x (Landrace x Large White) gilts, LL-F: Landrace x Large White gilts, DL-F: Duroc x (Landrace x Large White) gilts, PD-F: Pietrain x (Landrace x Duroc) gilts, PD-M: Pietrain x (Landrace x Duroc) entire males, PD-C: Pietrain x (Landrace x Duroc) surgically castrated males, PD-I: Pietrain x (Landrace x Duroc) immunocastrated males

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566 **Table 4** *Pearson correlation coefficients between intramuscular fat content of*  
567 *different muscles or anatomical positions for pigs from 70 to 120 kg live weight*  
568 *(n=153–155).*

	LD34LR	LD34VL	SM	BF	GM
LD89LR	0.74	0.78	0.63	0.60	0.70
LD34LR		0.83	0.56	0.53	0.62
LD34VL			0.65	0.62	0.66
SM				0.66	0.67
BF					0.63

All the correlations have a *P*-value <0.0001

LD89LR: between the 8<sup>th</sup> and 9<sup>th</sup> last ribs; LD34LR: between the 3<sup>rd</sup> and 4<sup>th</sup> last ribs;

LD34VL: between the 3<sup>rd</sup> and 4<sup>th</sup> lumbar *vertebrae*; SM: *Semimembranosus*; BF: *Biceps femoris*; GM: *Gluteus medius*.

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571 **Table 5** Pearson correlation coefficients between marbling and subcutaneous fat  
 572 thickness with intramuscular fat (IMF) in several muscles of pigs (n=148–153)<sup>1</sup>

Muscles <sup>2</sup>	Marbling (NPPC <sup>3</sup> )			Subcutaneous fat thickness (ruler)			
	LD89LR	LD34LR	LD34VL	LD89LR	LD34LR	LD34VL	Ham <sup>4</sup>
LD89LR	0.51	0.50	0.57	0.44	0.48	0.39	0.49
LD34LR	0.62	0.58	0.60	0.37	0.35	0.28	0.37
LD34VL	0.59	0.57	0.66	0.44	0.41	0.33	0.46
SM	0.33	0.37	0.41	0.46	0.44	0.42	0.48
BF	0.44	0.38	0.43	0.46	0.47	0.40	0.43
GM	0.45	0.47	0.50	0.54	0.54	0.52	0.56

573 <sup>1</sup> All the correlations have a P value <0.0001

574 <sup>2</sup> LD34LR: between the 3rd and 4th last ribs; LD34VL: between the 3rd and 4th lumbar  
 575 vertebrae; LD89LR: between the 8th and 9th last ribs; BF: *Biceps femoris*; GM: *Gluteus medius*;  
 576 SM: *Semimembranosus*

577 <sup>3</sup> National Pork Producers Council (NPPC, 1999) scale from 1: very low to 10: very high

578 <sup>4</sup> Fat thickness over the GM muscle

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582 **Table 6** *Statistical parameters of the prediction of intramuscular fat content (%)*  
583 *in pig's muscles from computed tomography images by means of partial least*  
584 *square regression (PLS).*

Predictors	Factors	Mean	SD	RMSEP <sub>CV</sub>	R <sup>2</sup>	RPD
<i>All the muscles</i>						
HU -50 to +120	2	1.78	0.72	0.66	0.17	1.09
<i>Loin muscles</i>						
HU -50 to +120	2	1.77	0.83	0.65	0.42	1.28
<i>Ham muscles</i>						
HU -50 to +120	1	1.79	0.58	0.56	0.07	1.03

HU: Hounsfield values; Factors: Number of PLS factors; RMSEP<sub>CV</sub>: Root mean square error of prediction obtained with cross-validation leave-one-out; RPD: Residual predictive deviation (s.d./RMSEP<sub>CV</sub>)

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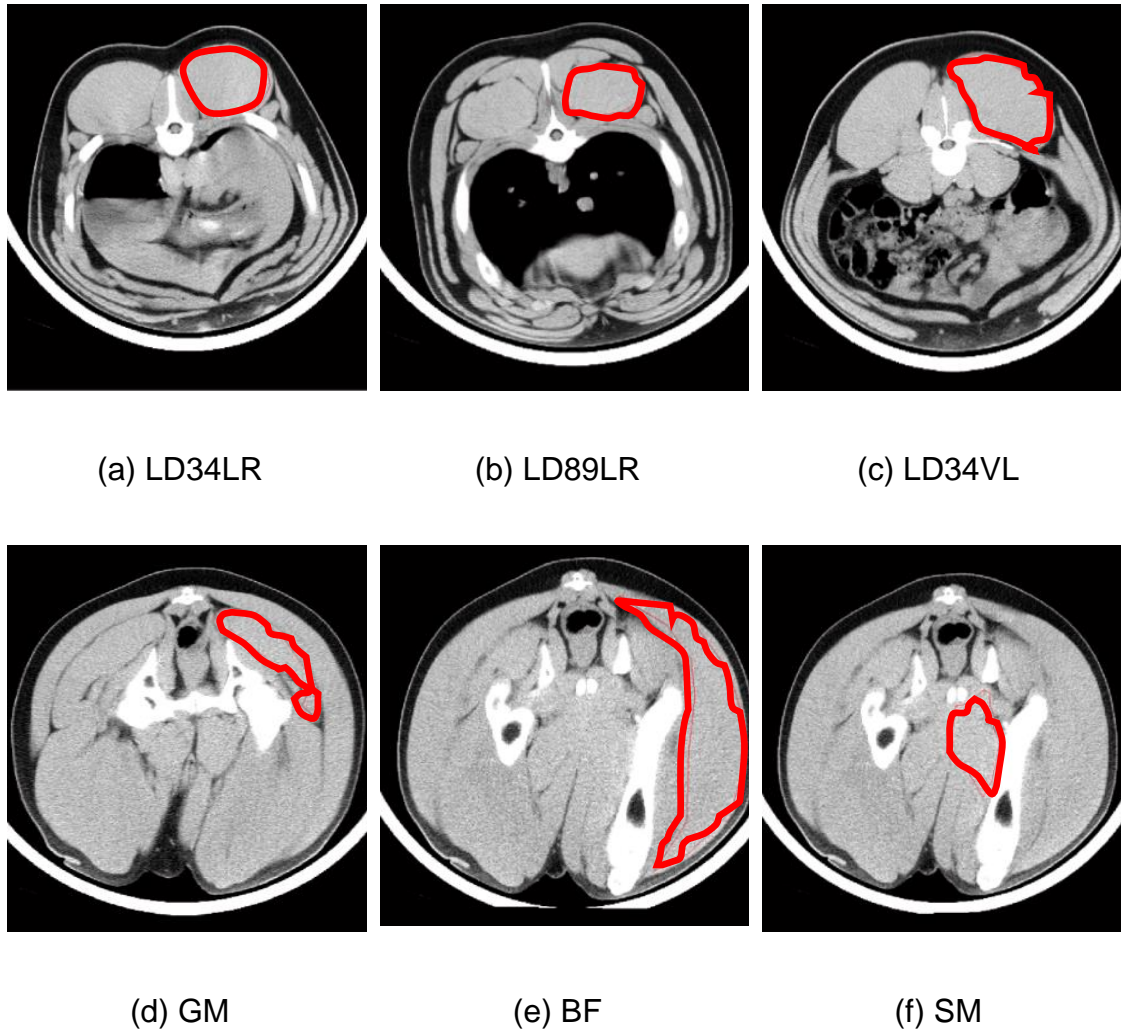
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591 **FIGURE CAPTIONS**

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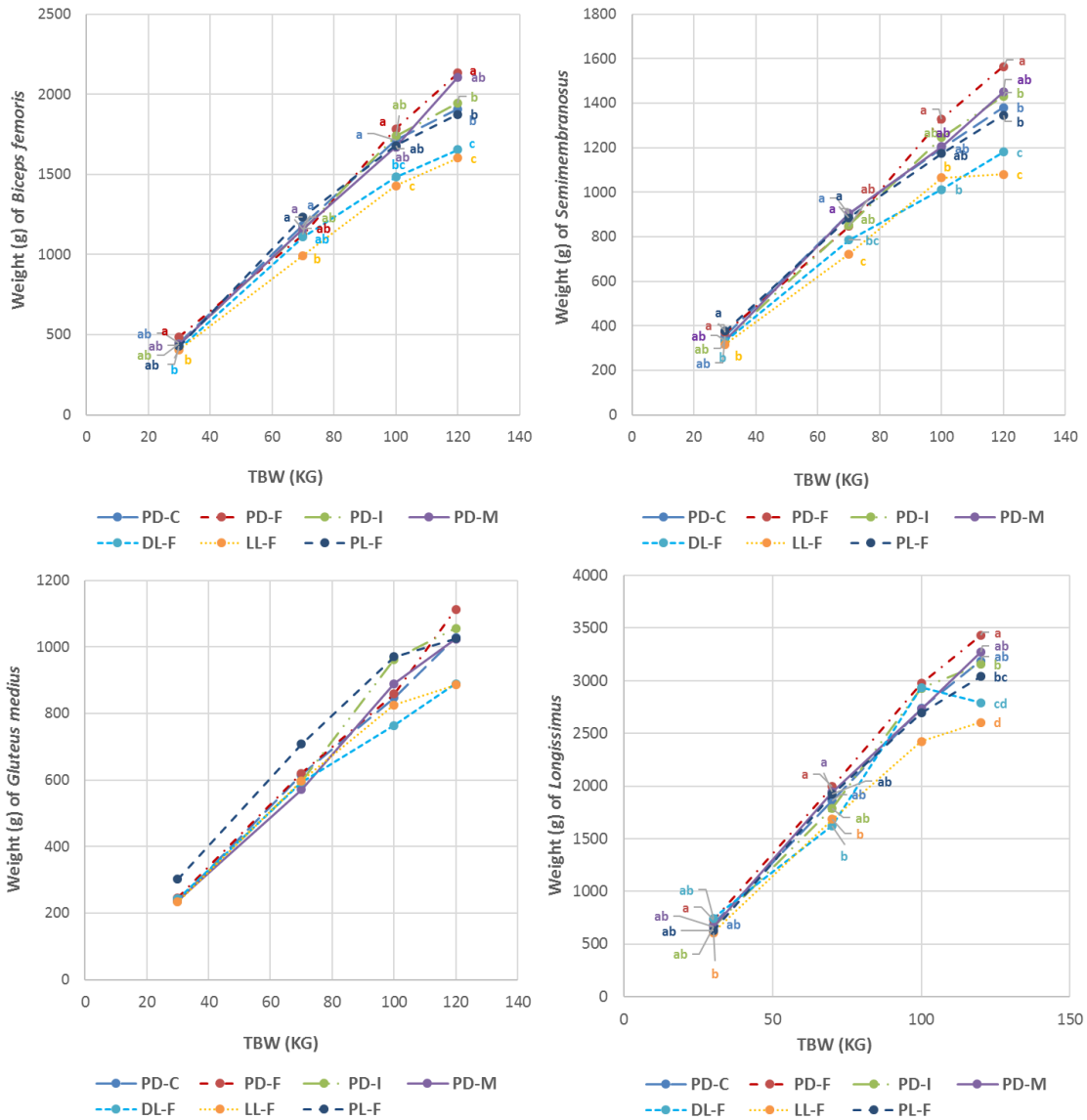


593 **Figure 1** Region of interest of the muscles and anatomical locations evaluated  
594 from computed tomography images of live pigs: (a) LD34LR, *longissimus*  
595 between the 3<sup>rd</sup> and 4<sup>th</sup> last ribs, (b) LD89LR, *longissimus* between the 8<sup>th</sup> and  
596 9<sup>th</sup> last ribs, (c) LD34VL, *longissimus* between the 3<sup>rd</sup> and 4<sup>th</sup> lumbar vertebra,  
597 (d) GM, *gluteus medius*, (e) BF, *biceps femoris* and (f) SM, *semimembranosus*

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602 **Figure 2** Trends of the weight of the muscles with the growth of the pig by  
 603 genotype-sex.

604 Footnote: PL-F : Pietrain x (Landrace x Large White) gilts, LL-F: Landrace x Large White gilts,  
 605 DL-F: Duroc x (Landrace x Large White) gilts, PD-F: Pietrain x (Landrace x Duroc) gilts, PD-M:  
 606 Pietrain x (Landrace x Duroc) entire males, PD-C: Pietrain x (Landrace x Duroc) surgically  
 607 castrated males, PD-I: Pietrain x (Landrace x Duroc) immunocastrated males; TBW: live target  
 608 body weight

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