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1 **A matter of body weight and sex type: pig carcass chemical composition and pork quality**

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13 **ABSTRACT**

14 This study compares minced carcass chemical composition and meat quality of castrated (CM),
15 immunocastrated (IM) and entire male (EM), and female (FE) pigs at 70, 100 and 120 kg target
16 body weights (TBW) (n=80; 20 per sex). Sex affected fat, protein, and moisture content of the
17 minced carcasses. Carcass fatty acid (FA) composition was affected by sex, with higher saturated
18 and monounsaturated FA content in CM than in FE, IM and EM, and higher polyunsaturated FA
19 in CM than in EM, with FE and IM in between. Except for intramuscular fat, which was higher
20 in CM than in FE and EM, no significant differences between sexes were found in meat quality.
21 TBW affected carcass chemical composition and some meat quality traits. An interaction between
22 sex and TBW was found with IM approaching EM or CM depending on TBW.

23

24 **Keywords:** minced carcasses, fat content, fatty acids, mineral content, immunocastration

25

26 **1. Introduction**

27 It is well known that nutritional and technological value of pork depend on the chemical
28 composition of the carcass, in particular its fat content and fatty acid profile, which are important
29 not only from the perspective of human health but also for meat processing and sensory properties
30 (Čandek-Potokar & Škrlep, 2012; Wood et al., 2004).

31 Focusing on the technological quality, fatty acid (FA) composition may affect the firmness of the
32 fat and the oxidative stability of the meat. The FA profile can be modified with diet (Duran-
33 Montgé et al., 2010; Pérez-Juan et al., 2010), and it is related to the level of carcass fatness as
34 well (Wood et al., 2008). Thus, differences among sex types known to differ in body and carcass
35 composition will lead to differences in the FA profile. A meta-analysis carried out by Pauly et al.
36 (2012) showed that saturated FA are lower and polyunsaturated FA are higher in entire males
37 than in castrated and immunocastrated males. Nevertheless, the changes associated to
38 immunocastration can be different depending on the fat depot (Poklucar et al., 2021). In addition,

39 differences in carcass chemical composition are expected to be greater at higher body weights
40 (Arthur et al., 2011; Mörlein and Tholen, 2015).

41 There is a tendency in the EU to abandon surgical castration of male pigs due to animal welfare
42 issues. Thus, if the use of this procedure is diminished or stopped and the production of entire
43 males or immunocastrates increases, final pork quality is likely to be altered. Nevertheless, for
44 both alternatives, published results on differences in meat quality are not always consistent
45 (Škrlep et al., 2020). Therefore, comparative studies with the sex types available in the market,
46 including female pigs, are essential. Since body composition changes with growth (Ruiz-
47 Ascacibar, 2018), studying the sex effect at distinct relevant stages of the pig growth adds to
48 understanding if the differences between sex types vary according to growth stage.

49 The objective of this study was to evaluate the gross chemical composition of the whole carcass
50 and some meat quality traits (i.e. pH, electrical conductivity, colour and intramuscular fat content)
51 of pigs from four sex types (castrated males, immunocastrated males, entire males, and females)
52 at three target body weights (TBW) i.e. at 70, 100 and 120 kg. Productive performance and the
53 characteristics of the carcass, main cuts and internal organs have been previously published
54 (Zomeño et al., 2022).

55

56 **2. Material and Methods**

57 The procedures used in this research were approved by the Committee of Ethics and Animal
58 Experimentation (CEEAA) of IRTA and Generalitat de Catalunya (DAAM Protocol number 5298).

59 *2.1. Animals and experimental design*

60 This study was carried out with 80 pigs of the same genetic type (Pietrain × (Landrace × Duroc))
61 from four different sex types (ST, n=20 each): surgically castrated males (CM), immunocastrated
62 males (IM), entire males (EM) and females (FE).

63 Thirty-two pigs (8 per sex) were reared in two collective pens (13.5 m²; 5.0 m length × 2.7 m
64 width) and slaughtered at 70 or 100 kg TBW. The remaining 48 pigs (12 per sex) were reared in

65 individual pens (3.0 m²; 2.5 m length × 1.2 m width) and slaughtered at 120 kg TBW. All pens
66 were equipped with a slatted floor surface, one drinking bowl and one free-access feeder. All the
67 pigs were fed *ad libitum* with the same commercial diet according to a two-phase feeding
68 programme containing 10.2 and 10.1 MJ net energy/kg, 18.0% and 17.0% CP and 0.91% and
69 0.90% digestible lysine, respectively. Surgical castration of male piglets was performed within
70 the first 7 days of age. For immunocastration the vaccine Improvac® (Zoetis, Alcobendas, Spain)
71 was administered twice: the first dose (V1) at 12 weeks of age and the second one (V2) at 18
72 weeks of age, at approximately 70 kg TBW. The vaccine was injected subcutaneously just behind
73 and below the base of the ear by technical staff in accordance with the manufacturer's instructions.
74 When pigs reached the desired TBW, they were slaughtered: 16 pigs at 70 kg (4 CM, 4 IM, 4 EM
75 and 4 FE), 17 pigs at 100 kg (4 CM, 4 IM, 5 EM and 4 FE), and 47 pigs at 120 kg (12 CM, 12
76 IM, 11 EM and 12 FE). This last TBW, 120 kg, was considered the most relevant one in this study
77 because it is closer to the commercial slaughter weight.

78 *2.2. Slaughtering and carcass measurements*

79 Slaughtering was carried out following standard procedures at the IRTA-Monells slaughterhouse
80 (located 300 m from the experimental farm) after animals were fasted for a minimum period of 8
81 hours. Pigs were stunned using 85% CO₂.

82 After evisceration and splitting of the carcass, backfat thickness (mm) was measured at the split
83 line of the left half carcass perpendicularly to the skin using a ruler at the following sites: between
84 the third and fourth lumbar vertebrae (Fat34LV); between the third and fourth last ribs (Fat34LR);
85 between the eighth and ninth last rib (Fat89LR); between the fifth and sixth thoracic vertebrae
86 (Fat56TV). Carcass lean meat percentage (LMP) was determined with Fat-O-Meat'er (FOM) II
87 (Frontmatec A/S, Smørum, Denmark) using the Spanish official FOM equation (European
88 Commission, 2020).

89 The carcasses (left and right half parts) were kept in a chilling room for approximately 24 hours
90 at 2-3°C and after that time they were weighted. Left half carcasses were used for meat quality

91 determinations (section 2.3). Afterwards, they were cut according to the European reference
92 method (Walstra & Merkus, 1995), and the proportion of the main cuts (ham, loin, shoulder, belly,
93 and tenderloin) with respect to carcass weight was calculated. See Zomeño et al. (2022) for a
94 more detailed description. Right half carcasses were stored at -20°C until the mincing process
95 and the subsequent chemical composition analysis (section 2.4).

96 *2.3. Meat quality analysis*

97 In all left half carcasses ($n=80$), the pH of the *Longissimus thoracis* (LT) muscle was determined
98 45 minutes after slaughter (pH 45min) and 24 hours post-mortem (pH 24h) at the level of the last
99 rib using a portable equipment with a Xerolyt electrode (Crison, Barcelona, Spain) with the
100 automatic temperature compensation function. The pH meter was calibrated with pH 7.0 and pH
101 4.0 buffers prior to measurements. At 24 hours post-mortem, electrical conductivity of the LT
102 muscle was recorded using the Pork Quality Meter (PQM-Kombi, Aichach, Germany) at the last
103 rib level. Colour was measured in the LT muscle, on the cross section at the level of the last rib
104 after 15-20 minutes of blooming. A subjective colour score was obtained using the Japanese scale
105 colour (Colour JSC: score 1–6, with 1: extremely pale to 6: extremely dark) (Nakai et al., 1975).
106 Scores were obtained from two experienced technicians, and the mean of their two scores was
107 recorded. An objective measurement (CIE, 2004) was performed using a Chroma-Meter 200
108 (Minolta Co., Ltd., Osaka, Japan) with standard illuminant D65, observation angle of 10° and an
109 aperture of 8 mm, and determining luminosity (L^*), redness (a^*) and yellowness (b^*) indexes.

110 Intramuscular fat of the LT muscle (IMF) was determined. Samples of 4-cm thickness were
111 obtained at the level of the last rib at 24 hours post-mortem, and then they were vacuum-packed
112 and stored at -20°C until the analysis. Intramuscular fat percentage was determined using Near
113 Infrared Transmittance equipment (Infratec 1265 Meat Analyzed, Tecator AB, Uppsala, Sweden)
114 scanned in the region between 800–1000 nm, in 2 nm increments. This equipment averaged the
115 results of 15 readings from each minced and homogenized sample of 100 g and it was calibrated
116 to predict total muscle fat triglycerides.

117 *2.4. Carcass chemical analysis*

118 First, the frozen entire right half carcasses (n=80) (including bones) were reduced into small
119 pieces using a cutting guillotine (Model D, Spain). The teeth were removed during this step. Then,
120 the different pieces were gradually placed in an industrial mincer (Grinder Cato-pa160, Spain)
121 and ground for three times using sieves with a smaller diameter each time. The minced sample
122 was thoroughly homogenized in a blender. Afterwards, samples (500 g) were collected, vacuum-
123 packed and stored at -20°C until further analysis.

124 In all carcasses (n=80), fat, protein, moisture, ash, Ca and P contents were determined in duplicate.
125 The fat content was determined by solvent extraction with petroleum ether (Soxtec 2050, Tecator,
126 Höganäs, Sweden) in a sample of around 5 g. The determination of the protein content was based
127 on the Kjeldahl total nitrogen content using a nitrogen/protein analyser (Büchi Distillation Unit
128 B-324 and Digester Unit B-414, Flawil, Switzerland) using a sample of 1.5 g. The moisture
129 content was determined by sample drying in an oven at 100°C to 105°C until constant weight in
130 a sample of approximately 10 g. The determination of the ash content was performed by heating
131 4 g of sample in a muffle oven at 550°C for 2 to 3 h. Finally, the Ca and P content were determined
132 by inductively coupled plasma-optical emission spectroscopy (ICP-OES; spectrophotometer
133 Optima 4300 DV, Perkin Elmer Inc., Wellesley, MA, USA). The sample (approximately 0.5 g)
134 was previously mineralized in a microwave (MARSXpress, CEM, Matthews, NC, USA).

135 Fatty acid (FA) profile was determined in 68 carcasses: 13 from those animals slaughtered at 70
136 kg (3-4 per sex type), 13 from those at 100 kg (3-4 per sex type), and 42 from those at 120 kg (9-
137 12 per sex type). Firstly, the fat extraction was performed using chloroform/methanol extraction
138 by Folch et al. (1957) from a sample of 10 g. Then, FA methyl esters were prepared using the
139 American Oil Chemists' Society (AOCS) Official Method Ce 2-66 and analysed in an Agilent
140 8860 Gas Chromatograph System (Agilent, Santa Clara, CA, United States) equipped with a
141 split/splitless injector. The separation of FA methyl esters was performed in a HP-88 column (Ref.
142 112-8867, Agilent, Santa Clara, CA, United States) ($60\text{ m} \times 0.25\text{ mm} \times 0.20\text{ }\mu\text{m}$) and detected
143 with a flame ionizing detector (FID) and a temperature gradient oven. For individual FA

144 identification a commercial reference (Supelco 37 Component FAME Mix Ref. CRM 47885,
145 Sigma-Aldrich, Seelze, Germany) was used.

146 *2.5. Statistical analysis*

147 Statistical analysis was performed with SAS v. 9.4 software (SAS Institute Inc., Cary, NC, USA).
148 Data were analysed using a generalized linear mixed model (GLIMMIX procedure) including
149 TBW (70, 100, and 120 kg), ST (CM, IM, EM, and FE), and their interaction as fixed effects. For
150 those variables more dependent on body weight (i.e. cold carcass weight, subcutaneous fat
151 thicknesses, fat, protein, and moisture content of the minced carcass, and IMF content),
152 heteroscedasticity was corrected by using the weighted least square approach (Font-i-Furnols et
153 al., 2015). The SLICEDIFF option was used for the comparison of LSMEANS. Tukey test was
154 used to compare the adjusted LSMEANS values at the 0.05 significance level.

155 Principal components analysis (PCA) was applied to obtain a multidimensional view of the
156 studied traits' interrelationship. Therefore, the FACTOR procedure of SAS was used applying a
157 mean and centring pre-processing. For a more global view of the characteristics of carcass and
158 meat quality, FA percentages (%) as well as data of estimated carcass lean meat (%) and the
159 proportion (g/kg) of the main cuts (ham, loin, shoulder, belly and tenderloin) evaluated at the
160 same animals (Zomeño et al., 2022) were included in this analysis. A first PCA was conducted
161 including those carcasses with all data (n=68), and a second PCA including those from animals
162 slaughtered at 120 kg and with all data (n=42).

163

164 **3. Results and discussion**

165 *3.1. Carcass composition*

166 Results from carcass characteristics and their proximate composition are presented in Table 1 and
167 Supplementary Figures 1 and 2. Sex had a significant effect on carcass weight and subcutaneous
168 fat thicknesses measured at all sites, and this effect was constant across the 3 TBW since no
169 significant TBW × ST interaction was found. Castrated males and FE presented heavier carcasses

170 than IM, with EM carcasses being in between (Table 1 and Supplementary Figure 1). Regarding
171 the subcutaneous backfat, carcasses of CM and FE showed greater thicknesses than IM in the ham
172 area (Fat34LV). In the loin area, however, CM showed higher values than IM and EM at the point
173 between the 3rd and 4th last ribs (Fat34LR), and higher than EM between the 8th and 9th last ribs
174 (Fat89LR) and the 5th and 6th thoracic vertebrae (Fat56TV) (Table 1 and Supplementary Figure
175 1). Female carcasses had intermediate fatness levels at the three anatomical locations of the loin
176 area. The differences obtained are in line with those observed in other anatomical fat positions in
177 the same animals (Zomeño et al., 2022) and confirm a different carcass fat deposition in IM pigs;
178 they were more like EM in the ham area and more like CM in the loin area. These findings agree
179 with previous studies comparing body and carcass composition of these four sexes (Fàbrega et
180 al., 2010; Gispert et al., 2010). For some variables, especially those related to fat content, the
181 interaction was not significant, although we expected it to be because of the inclusion of IM pigs
182 in the trial. The lack of significance for the interaction can be due to the fact that the interaction
183 does not exist or because of the small number of animals at 70 and 100 kg TBW. Despite this low
184 animal number at 70 and 100 kg TBW, the outcome provided is novel and valuable.

185 Significant differences among sexes were observed for fat, protein and moisture content of the
186 minced carcasses while the ash and mineral content (P and Ca) did not change (Table 1).
187 Carcasses of CM had, overall, a higher fat content than those of FE and these, in turn, had a higher
188 content than EM. Immunocastrated carcasses were leaner than CM and similar to FE and EM
189 (Supplementary Figure 2). For protein content, carcasses of EM and FE had higher values than
190 those of CM, with IM being in between (Table 1 and Supplementary Figure 2). Moisture content
191 of carcasses from different sexes changed with TBW (TBW × ST interaction P = 0.001). In detail,
192 EM and IM showed a higher moisture content than CM at 70 kg and 100 kg, and at 120 kg EM
193 had the highest value, FE and IM had intermediate values, and CM the lowest value. The
194 differences obtained for fat and moisture content across sexes are in line with the allometric
195 growth of these components estimated by Zomeño et al. (2016) in the same four sexes. These
196 authors did not find any difference in the allometric coefficients of protein, but Schinckel et al.

197 (2008) reported higher values in FE than in CM, in line with our results. Zomeño et al. (2016)
198 obtained a similar growth for ash content between sexes, in agreement with the present results;
199 although they estimated different allometric coefficients for Ca and P that were not corroborated
200 in our study.

201 Carcass weight, subcutaneous fat thickness and fat content of minced carcasses significantly
202 increased with increasing TBW (Table 1 and Supplementary Figures 3 and 4). On the contrary,
203 protein, moisture, ash and P content decreased. These findings were expected and agree with the
204 evolution of carcass composition with animal age and body weight (Arthur et al., 2011; Schinckel
205 et al., 2008; Ruiz-Ascacibar, 2018).

206 Table 2 shows FA composition of minced carcasses according to TBW and ST. Significant
207 differences among sexes were observed for all the FA groups and for several individual FA. The
208 total content of saturated (SFA) and monounsaturated (MUFA) FA was higher in CM than in IM,
209 EM and FE and these differences were constant across TBW (Table 2 and Supplementary Figure
210 5). These results were caused by the differences observed in the palmitic (C16:0) and stearic
211 (C18:0) acid total content, and by those observed in the oleic acid (C18:1 n-9 cis), which are the
212 most abundant individual SFA and MUFA in the pig minced carcass, respectively. Other
213 individual SFA and MUFA were affected by the ST as well (Table 2). For instance, myristic
214 (C14:0), margaric (C17:0), palmitoleic (C16:1), and gadoleic (C20:1) acids. In addition, the total
215 content of polyunsaturated FA (PUFA) was higher in CM compared to EM, with IM and FE in
216 between (Table 2 and Supplementary Figure 5). This was mainly due to the higher total content
217 of linoleic acid (C18:2 n-6) in CM than in EM. The total content of n-6 and n-3 FA was higher
218 in CM than in EM as well. Likewise other individual PUFA such as eicosadienoic (C20:2) and
219 eicosatrienoic (C20:3) were affected by the ST, whereas linolenic (C18:3 n-3) and arachidonic
220 (C20:4 n-6) acids were not altered. Finally, the PUFA/SFA ratio was lower in CM than in the
221 other three ST, whereas the n-6/n-3 ratio was not altered. Overall, these differences are consistent
222 with those observed for carcass fat content (Table 1) and can be explained by the fat saturation
223 (corresponding to a higher de novo synthesis of SFA and MUFA) and a more diluted effect on

224 PUFA content with increasing fat content (Mauvoisin and Mounier, 2011; Wood et al., 2008).
225 The FA composition of the minced carcass can give an overview of the global nutritional
226 characteristics of the carcass. However, most published studies have focused on FA composition
227 of specific porcine tissues, and there is limited information on the FA of the whole pig carcass.
228 Moreover, to our knowledge, no studies have been published comparing the whole carcass FA
229 content between sexes, which makes this study novel and unique. Previous investigations in the
230 subcutaneous fat (loin area) and LT muscle reported lower SFA and higher PUFA percentages in
231 EM than CM (Mackay et al., 2013; Pauly et al., 2012; Poklukar et al., 2021). Font-i-Furnols et al.
232 (2012) found higher MUFA percentage in the subcutaneous fat (ham area) of FE than IM. These
233 findings agree with our results when expressing FA as percentages, with more monounsaturated
234 fat in CM and FE than in IM and EM (47.1% and 46.7% vs. 45.3% and 44.7%) and less
235 polyunsaturated fat in CM than in EM, IM and FE (16.9% vs. 20.2%, 19.6% and 18.9%,
236 respectively). Nevertheless, in our study, IM showed similar values to EM and FE for PUFA
237 percentage and to EM for MUFA percentage, which is consistent with results of Mackay et al.
238 (2013), but not with the other above-mentioned studies. Therefore, the discrepancies observed
239 between studies could be explained by a different response of FA composition depending on the
240 anatomical location of the sample analysed (e.g. subcutaneous fat vs. intramuscular fat vs. whole
241 carcass fat). Indeed, FA profile varies according to fat tissue location (Daza et al. 2017). Recently,
242 Poklukar et al. (2021) showed that immunocastration changed FA profile more slowly at the
243 withers than at the last rib, and more rapidly in the LT muscle (IM were closer to CM than to
244 EM).

245 Target body weight affected FA total content (Table 2 and Supplementary Figure 6). The content
246 of SFA and MUFA was higher in pigs slaughtered at 120 kg than at 70 and 100 kg TBW. The
247 content of PUFA, n-6 and n-3 was higher in pigs slaughtered at 100 kg and 120 kg than in pigs
248 slaughtered at 70 kg TBW. These results are coherent with the increase in carcass fat content with
249 increasing animal age and body weight.

250 When TBW and ST were considered together, SFA total content was 1.4 times higher in CM than
251 in IM and EM at 70 kg, but at 100 kg it was 1.34 times higher than IM and 1.49 times higher than
252 EM, and at 120 kg, 1.20 and 1.55 times higher than IM and EM, respectively (Table 2). Thus,
253 although no significant interaction $TBW \times ST$ was found, differences in SFA content between
254 CM and EM were similar to those between CM and IM before the second vaccination, but the
255 difference in SFA content was higher between CM and EM than between CM and IM, indicating
256 an increase in the saturation of fat of IM pigs after the second vaccination, and approaching those
257 of CM in agreement with what has been published in literature (Škrlep et al., 2020). A similar
258 pattern was found for MUFA since at 70 kg CM had 1.35 and 1.47 times higher total content than
259 IM and EM, respectively, while at 120 kg CM had 1.25 and 1.59 times higher content than IM
260 and EM, respectively. As expected, the opposite situation was found for PUFA because CM had
261 1.05, 1.17 and 1.16 times higher total content than IM at 70, 100 and 120 kg, respectively, while
262 they had 1.17, 1.13 and 1.27 times higher total content than EM at 70, 100 and 120 kg,
263 respectively. These findings can be associated to the differences in carcass fat content between
264 sexes (Škrlep et al., 2020; Gispert et al., 2010).

265 3.2. Meat quality

266 Meat quality traits measured in the LT muscle are shown in Table 3. Intramuscular fat percentage
267 was higher in CM than in FE and EM, with IM in between (Table 3 and Supplementary Figure
268 7). This result agrees with those observed for the subcutaneous fat thickness in the loin area
269 (Fat89LR and Fat56TV) and the fat content of minced carcass in the present work. A significant
270 $TBW \times ST$ interaction was found for electrical conductivity, although the differences among LS-
271 means were not relevant in terms of technological quality. In fact, the values obtained for
272 electrical conductivity were lower than 6, which is a common cut-off value used to classify meat
273 as PSE (Guàrdia et al., 2004). Redness index was higher in CM than in FE, but these differences
274 were only found at 70 kg TBW ($TBW \times ST$ interaction). Therefore, except for IMF percentage,
275 there were no major differences observed in meat quality between sexes, which agrees with

276 literature (see meta-analyses of Pauly et al. (2012) and Trefan et al. (2013) or the review of Škrlep
277 et al. (2020)) usually reporting inconsistent results on the effect of sex on pork quality.

278 The pH value at 45 minutes and at 24 hours post-mortem was affected by the TBW, with higher
279 values in pigs of 70 kg than in pigs of 100 and 120 kg TBW group. Consistent with pH values,
280 lightness was higher at 120 kg than at 70 kg (Table 3 and Supplementary Figure 8). Higher redness
281 at 100 and 120 kg than at 70 kg can partly be related to higher luminosity and partly to higher age
282 of pigs. However, as already explained for the effect of sex, the differences in meat quality were
283 not consistent.

284 *3.3. Relationship between carcass composition and meat quality*

285 Figure 1 shows the results of the principal component analysis including all samples. The first
286 principal component (PC1) (horizontal axis) explains 30.2% and the second principal component
287 (PC2) (vertical axis) explains 17.1% (left plot) of the total variation. Fatty acids included in the
288 PCA are expressed in percentage in order to differentiate the amount of fat with its quality and
289 explore differences in the proportion of the different FA among sexes and weights. The PC1 is
290 related to carcass lean (LMP) and protein and moisture content (minced carcass) on the right side
291 (positive correlation to PC1). On the left side (negative correlation to PC1), variables related to
292 fat (minced carcass fat content, subcutaneous fat thickness, and IMF) and MUFA percentage are
293 placed. In addition, the proportions of ham, shoulder and tenderloin are placed on the right side
294 whereas those of loin and belly as well as carcass weight are on the left side (data from Zomeño
295 et al., 2022). The PC2 is mainly explained by the SFA percentage on the negative side (bottom)
296 and by the PUFA, n-6 and n-3 on the positive side (top). Considering the experimental groups,
297 EM are placed on the right side of PC1 because they are associated with higher carcass leanness
298 and protein content. However, CM are placed on the left side of PC1 because they are associated
299 with higher carcass fat content. Females and IM are in the middle part, and thus, not related to
300 any of the PCs considered because they have intermediate levels of fatness. The TBW groups are
301 separated by the PC 1 and 2. The most evident is the group slaughtered at 120 kg, which was
302 placed on the negative side of PC1; it is associated with higher carcass fatness, as expressed by

303 several variables such as fat thickness and IMF. The group slaughtered at 100 kg was positioned
304 on the positive side of PC1 and PC2, and closer to PUFA. The group slaughtered at 70 kg is placed
305 on the right side (lower part) and more related to leaner carcasses. The right plot of Figure 1 shows
306 the results when plotting the third PC (vertical axis). Although this PC explains only 8.2% of
307 variation, it clearly separates FE (placed on the negative side) from IM (placed on the positive
308 side). In this case, IM are positively related to SFA content, Ca and P content, and lighter meat
309 colour, and FE with darker meat colour, carcass MUFA content and ham proportion. Overall,
310 these results corroborate well with those obtained when the traits were analysed individually (see
311 Tables 1 and 2 of this paper).

312 Figure 2 shows the results of the PC analysis considering only the group slaughtered at 120 kg
313 TBW. In this case, PC1 is associated with PUFA and n-6 percentages that are placed on the
314 positive right and with leaner carcasses, carcass protein and moisture content, while SFA is placed
315 on the negative side close to fat variables. The PC2 explains mainly the variables related to muscle
316 colour and MUFA percentage. Males are clearly separated by PC1, with EM on the right side,
317 CM on the left one, and IM in an intermediate position although closer to CM. Females are
318 represented by the PC2.

319 Looking at the two PCA analyses, it can be seen that EM are characterised by leaner carcasses at
320 all three TBW and, in addition, by having more n-6 PUFA and greater ham, shoulder and
321 tenderloin yields at 120 kg. Castrates are characterised by fatter carcasses with more SFA and
322 greater belly and loin yields at all three TBW. Immunocastrates and FE are not separated by their
323 fat/lean content when all three TBW are included while, at 120 kg, differences among all sexes
324 are clearer. In the case of IM, this is probably explained by the change in fatness after the second
325 dose of the vaccine, since they are closer to EM until that moment, and thereafter they rapidly
326 become fatter and approach CM (Batorek et al., 2012; Poulsen Nautrup et al., 2018). While in the
327 case of FE, their fatness being intermediate between CM and EM (Gispert et al. 2010) explains
328 their position.

329

330 **4. Conclusions**

331 The sex of the pigs does not seem to affect technological meat quality characteristics. However,
332 sex affects intramuscular fat, which is important for the eating quality of the meat. Moreover, pig
333 sex and their body weight proved to be significant factors affecting the content of the main
334 chemical constituents of the carcass, and these modifications lead to important changes of FA
335 profile, which can affect at the subsequent processing and shelf life of the product, because the
336 higher the SFA, like in CM, the better the processing and the lower the oxidation. While FE, CM
337 and EM presented more consistent characteristics across the finishing body weights, IM traits
338 changed more due to the vaccination pattern mainly in terms of carcass fatness and FA
339 composition. Therefore, the interaction between pig sex and body weight needs to be considered
340 when producing carcasses, cuts, or meat products with certain required characteristics. Further
341 studies with a higher number of animals would be necessary to corroborate these findings.

342

343 **Author's contribution**

344 Cristina Zomeño: Formal analysis, writing - original draft, writing - review & editing.

345 Marina Gispert: Conceptualization, data curation, formal analysis, funding acquisition,
346 investigation, writing - original draft, writing - review & editing.

347 Marjeta Čandek-Potokar: Formal analysis, writing - original draft, writing - review & editing.

348 Daniel Mörlein: Formal analysis, writing - original draft, writing - review & editing.

349 Maria Font-i-Furnols: conceptualization, data curation, formal analysis, funding acquisition,
350 investigation, methodology, writing - original draft, writing - review & editing.

351

352 **Ethical statement**

353 The procedures used in this research were approved by the Committee of Ethics and Animal
354 Experimentation (CEEA) of IRTA and Generalitat de Catalunya (DAAM Protocol number 5298).

355

356 **Declaration of competing interest**

357 The authors declare no conflict of interest associated with this research.

358

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366

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464 TABLES

465

466 **Table 1.** Carcass traits and chemical composition of minced carcass (LSMEANS) of pigs according to sex type (ST) and slaughtered at three target body weights
467 (TBW)

TBW	70 kg				100 kg				120 kg				P-value			RMSE	
	ST	CM	IM	EM	FE	CM	IM	EM	FE	CM	IM	EM	FE	TBW	ST		TBW × ST
<i>Pigs (n)</i>		4	4	4	4	4	4	5	4	12	12	11	12				
Cold carcass weight (kg) ^{av}		56.26	53.25	56.44	57.19	80.99	78.80	81.17	81.65	96.50	94.65	94.69	96.71	<0.001	0.004	0.632	0.485
Carcass fatness (mm)																	
Fat34LV ^{bv}		20.75	15.00	21.00	20.25	29.75	23.50	30.25	24.40	29.25	28.25	28.83	24.82	<0.001	0.003	0.132	2.057
Fat34LR ^{aw}		13.50	10.25	12.50	12.75	21.75	16.75	15.80	15.75	23.42	21.00	18.00	19.83	<0.001	0.015	0.521	1.973
Fat89LR ^{ax}		17.25	12.75	13.00	16.75	23.50	21.25	20.20	19.25	28.33	26.33	23.09	24.67	<0.001	0.024	0.559	1.984
Fat56TV ^{ax}		22.75	21.75	21.00	23.50	30.25	26.75	26.00	25.50	34.66	31.92	27.62	31.42	<0.001	0.001	0.255	1.812
Chemical composition of minced carcasses																	
Fat (g/100 g) ^{ay}		15.37	11.02	11.71	14.65	20.17	15.22	14.09	16.47	24.94	20.50	16.62	20.50	<0.001	<0.001	0.157	1.658
Protein (g/100 g) ^{yz}		18.69	18.13	19.19	18.83	17.70	18.99	18.86	18.94	17.29	17.54	18.51	18.30	<0.001	<0.001	0.168	0.869
Moisture (g/100 g)		60.42 ^c	65.53 ^a	64.03 ^{ab}	61.05 ^{bc}	57.12 ^b	61.63 ^a	63.09 ^a	60.86 ^{ab}	53.78 ^c	56.15 ^b	60.95 ^a	57.89 ^b	<0.001	<0.001	0.001	1.433
Ash (g/100 g) ^δ		3.34	3.21	3.38	3.41	2.90	3.31	3.27	2.92	2.96	3.08	3.18	2.90	0.014	0.258	0.644	0.346
Ca (mg/g)		9.56	8.55	9.02	9.92	9.80	9.75	7.27	8.56	7.98	9.22	7.70	7.35	0.091	0.318	0.391	1.978
P (mg/g) ^δ		6.14	5.66	5.90	6.35	5.95	6.12	4.90	5.51	5.09	5.75	5.11	4.94	0.018	0.435	0.426	0.959

468 CM = surgically castrated males; IM = immunocastrated males; EM = entire males; FE = females; RMSE = root mean square error; Fat34LV = fat thickness between the 3rd and 4th lumbar
469 vertebrae; Fat34LR = fat thickness between the 3rd and 4th last ribs; Fat89LR = fat thickness between the 8th and 9th last ribs; Fat56TV = fat thickness between the 5th and 6th thoracic vertebrae.

470 ^{a,b,c} different superscripts within row indicate significant differences among LSMEANS of TBW×ST (P-value ≤ 0.05) (Tukey test).

471 ^{α,β,γ,δ} indicate significant differences among LSMEANS of TBW (P-value ≤ 0.05, Tukey test): ^α 70 kg < 100 kg < 120 kg; ^β 70 kg < 100 kg ≈ 120 kg; ^γ 70 kg ≈ 100 kg > 120 kg; ^δ 70 kg > 120 kg.

472 ^{v,w,x,y,z} indicate significant differences among LSMEANS of ST (P-value ≤ 0.05, Tukey test): ^v CM ≈ FE > IM; ^w CM > IM ≈ EM; ^x CM > EM; ^y CM > FE > EM; ^z CM < EM ≈ FE.

473 **Table 2.** Carcass fatty acid composition (mg/100 g of minced carcass) (LSMEANS) of pigs according to sex type (ST) and slaughtered at three target body
 474 weights (TBW)

TBW ST	70 kg				100 kg				120 kg				P-value			RMSE
	CM	IM	EM	FE	CM	IM	EM	FE	CM	IM	EM	FE	TBW	ST	TBW × ST	
<i>Pigs (n)</i>	3	3	3	4	3	3	4	3	12	10	9	11				
Σ SFA ^{av}	5625	3949	3877	5432	6774	5025	4817	5136	9308	7722	5999	6806	<0.001	<0.001	0.294	1355.2
C10:0	7	7	5	9	8	5	7	9	13	10	10	23	0.750	0.252	0.965	14.3
C12:0	10	9	6	9	11	5	11	13	16	11	11	12	<0.001	0.037	0.170	3.0
C14:0	182	125	126	171	210	161	164	180	290	225	196	222	<0.001	<0.001	0.571	40.1
C15:0	9	13	9	9	4	6	9	8	8	8	7	7	0.002	0.467	0.110	1.8
C16:0	3570	2507	2450	3376	4291	3179	3087	3295	5805	4815	3738	4313	<0.001	<0.001	0.323	794.4
C17:0	60	52	34	43	48	45	45	47	57	47	39	46	0.946	0.002	0.411	9.5
C18:0	1744	1235	1223	1783	2195	1618	1496	1565	3074	2574	1969	2134	<0.001	0.002	0.228	487.8
C20:0	23	8	15	22	17	9	8	26	48	35	28	36	<0.001	0.012	0.398	11.6
Σ MUFA ^{av}	7030	5207	4780	6705	9220	6393	6304	7310	12335	9838	7742	9796	<0.001	<0.001	0.438	1513.6
C16:1	412	285	273	362	455	343	349	398	598	488	410	510	<0.001	<0.001	0.804	73.3
C17:1	40	37	23	31	40	33	35	39	49	37	31	41	0.024	<0.001	0.330	7.4
C18:1 n-9 cis	6431	4772	4386	6155	8556	5901	5795	6718	11431	9121	7120	9035	<0.001	<0.001	0.409	1411
C18:1 n-9 trans	39	34	26	50	37	29	27	28	48	39	31	37	0.015	0.002	0.074	8.8
C20:1	108	79	72	105	132	87	98	128	209	153	151	172	<0.001	0.005	0.883	33.9
Σ PUFA ^{βw}	2497	2386	2137	2508	3582	3064	3183	3436	4079	3509	3201	3615	<0.001	0.027	0.755	462.8
C18:2 n-6 cis	2187	2121	1879	2202	3129	2696	2784	2982	3563	3081	2788	3139	<0.001	0.032	0.724	403.5
C18:3 n-3	119	118	102	122	213	177	181	190	221	190	163	187	<0.001	0.064	0.760	32.7
C20:2	107	93	90	104	128	92	106	134	170	136	142	158	<0.001	0.013	0.919	24.6
C20:3 n-9	59	44	52	61	89	77	90	96	93	76	78	98	<0.001	0.041	0.944	17.2
C20:4 n-6	14	15	11	15	24	18	21	26	32	26	27	29	<0.001	0.169	0.824	5.5
Σ n-3 ^{βw}	123	117	102	121	212	177	180	190	221	189	162	186	<0.001	0.047	0.811	32.39
Σ n-6 ^{βw}	2201	2130	1889	2217	3152	2714	2805	3007	3595	3106	2815	3168	<0.001	0.032	0.728	407.57
Σ PUFA/ Σ SFA ^{γx}	0.437	0.618	0.551	0.461	0.545	0.610	0.662	0.683	0.447	0.461	0.539	0.538	<0.001	0.006	0.052	0.076
Σ n-6/ Σ n-3 ^δ	19.03	18.02	18.70	18.28	14.83	15.27	15.46	15.85	16.25	16.31	17.57	16.98	<0.001	0.455	0.994	2.108

475 CM = surgically castrated males; IM = immunocastrated males; EM = entire males; FE = females; RMSE = root mean square error; SFA = saturated fatty acids; MUFA = monounsaturated fatty
 476 acids; PUFA = polyunsaturated fatty acids.

477 ^{a,b,c} different superscripts within row indicate significant differences among LSMEANS of TBW×ST (P-value ≤ 0.05) (Tukey t-test); ^{A,B,C} P-value ≤ 0.10 (Tukey test).
 478 ^{α,β,γ,δ} indicate significant differences among LSMEANS of TBW for fatty acid groups and ratios (P-value ≤ 0.05, Tukey test): ^α 70 kg ≈ 100 kg < 120 kg; ^β 70 kg < 100 kg ≈ 120 kg; ^γ 70 kg ≈
 479 120 kg < 100 kg; ^δ 70 kg > 100 kg ≈ 120 kg.
 480 ^{v,w,x} indicate significant differences among LSMEANS of ST for fatty acid groups and ratios (P-value ≤ 0.05, Tukey test): ^v CM > IM ≈ EM ≈ FE; ^w CM > EM; ^x CM < IM ≈ EM ≈ FE.
 481

482 **Table 3.** Meat quality traits of *Longissimus thoracis* (LSMEANS) of pigs according to sex type (ST) and slaughtered at three target body weights (TBW)

TBW	70 kg				100 kg				120 kg				P-value			RMSE	
	ST	CM	IM	EM	FE	CM	IM	EM	FE	CM	IM	EM	FE	TBW	ST		TBW × ST
<i>Pigs (n)</i>		4	4	4	4	4	4	5	4	12	12	11	12				
pH 45min ^α		6.60	6.75	6.71	6.82	6.52	6.63	6.51	6.40	6.63	6.67	6.54	6.70	0.024	0.491	0.568	0.208
pH 24h ^β		5.63	5.82	5.66	5.71	5.59	5.58	5.56	5.59	5.60	5.54	5.58	5.54	0.002	0.822	0.352	0.128
EC 24h (mS)		3.28	3.96	3.91	4.48	3.72	3.58	4.16	4.75	4.18	3.79	4.45	3.82	0.719	0.034	0.038	0.666
Colour JSC		2.38	2.63	2.63	2.25	2.89	2.25	2.00	3.13	2.63	2.33	2.50	2.54	0.891	0.431	0.122	0.580
L* (luminosity) ^γ		47.77	47.10	48.25	47.29	47.06	49.64	49.82	47.99	48.69	49.73	49.58	49.05	0.017	0.185	0.719	1.963
a* (redness) ^δ		7.17 ^a	5.74 ^{ab}	6.62 ^{ab}	5.31 ^b	7.20	6.75	7.52	7.66	6.94	7.20	6.62	7.29	0.002	0.389	0.021	0.899
b* (yellowness)		2.02	1.52	2.13	1.69	1.55	1.92	2.63	1.85	1.77	2.13	1.74	1.63	0.782	0.469	0.495	0.862
IMF (%) ^v		1.35	1.03	1.09	1.03	1.53	1.11	1.04	1.04	1.40	1.22	1.08	1.10	0.688	0.013	0.977	0.580

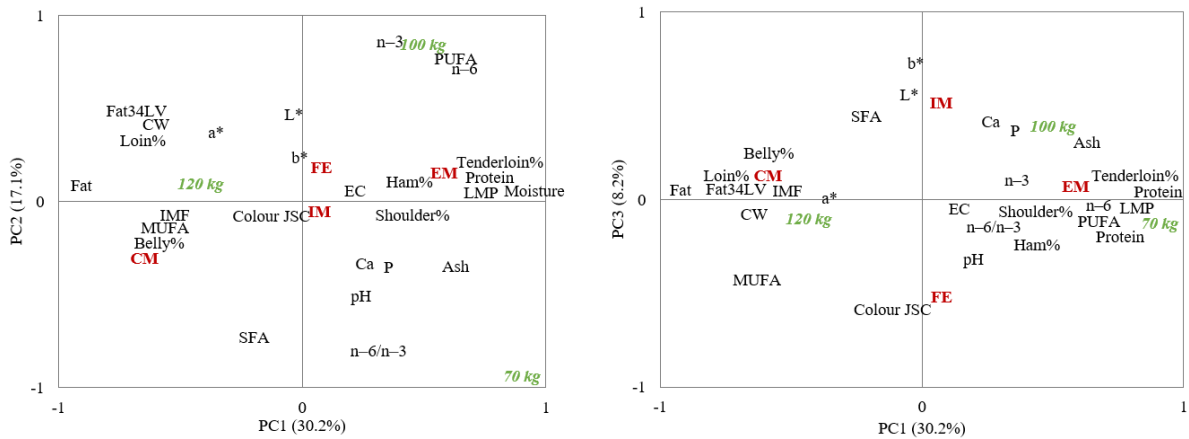
483 CM = surgically castrated males; IM = immunocastrated males; EM = entire males; FE = females; RMSE = root mean square error; EC = Electrical conductivity; JSC = Japanese scale of colour
 484 (1: pale, 6: extremely dark); IMF = intramuscular fat.

485 ^{a,b} different superscripts within row indicate significant differences among LSMEANS of TBW×ST (P-value ≤ 0.05) (Tukey test).

486 ^{α,β,γ,δ} indicate significant differences among LSMEANS of TBW (P-value ≤ 0.05, Tukey test): ^α 70 kg > 100 kg; ^β 70 kg > 100 kg ≈ 120 kg; ^γ 70 kg < 120 kg; ^δ 70 kg < 100 kg.

487 ^v indicates significant differences among LSMEANS of ST (P-value ≤ 0.05, Tukey test): CM > EM ≈ FE.

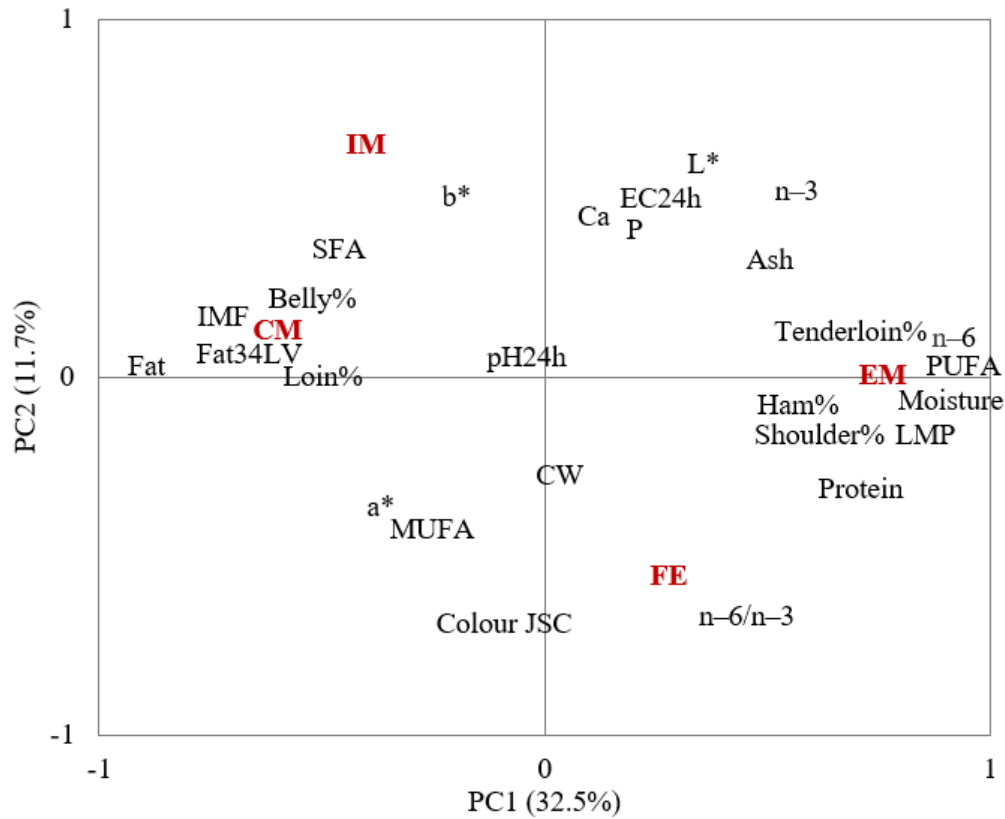
488 **FIGURE CAPTIONS**



489

490 **Figure 1.** Principal component (PC) analysis with all carcasses (n=68): PC1 and 2 (left plot), PC1
 491 and 3 (right plot). In brackets the variance accounted for each axis. Correlations between variables
 492 and factors (black colour) and coordinates of the experimental groups (red colour and bold letters
 493 = sex type; green colour, bold and cursive letters = target body weight). Carcass quality: CW (cold
 494 carcass weight), Fat34LV (fat thickness between the 3rd and 4th lumbar vertebrae), SFA
 495 (saturated fatty acids percentage); MUFA (monounsaturated fatty acids percentage), PUFA
 496 (polyunsaturated fatty acids percentage), LMP (estimated carcass lean percentage) and proportion
 497 of cuts with respect to CW: Ham%, Loin%, Shoulder%, Belly% and Tenderloin% (Zomeño et
 498 al., 2022). Meat quality of *Longissimus thoracis* at 24 hours *post-mortem*: pH, EC (electrical
 499 conductivity), Colour JSC (Japanese scale of colour), L*(luminosity), a*(redness), b*
 500 (yellowness), and IMF (intramuscular fat percentage). CM: surgically castrated males; IM:
 501 immunocastrated males; EM: entire males; FE: females.

502



503

504 **Figure 2.** Principal component (PC) analysis with carcasses from animals slaughtered at 120 kg

505 target body weight (n=42). In brackets the variance accounted for each axis. Correlations between

506 variables and factors (black colour) and coordinates of the different sex groups (red colour and

507 bold letters). Carcass quality: CW (carcass weight), Fat34LV (fat thickness between the 3rd and

508 4th lumbar vertebrae), SFA (saturated fatty acids percentage); MUFA (monounsaturated fatty

509 acids percentage), PUFA (polyunsaturated fatty acids percentage), LMP (estimated carcass lean

510 percentage) and proportion of cuts with respect to CW: Ham%, Loin%, Shoulder%, Belly% and

511 Tenderloin% (Zomeño et al., 2022). Meat quality of *Longissimus thoracis* at 24 hours post-

512 mortem: pH, EC (electrical conductivity), Colour JSC (Japanese scale of colour), L*(luminosity),

513 a*(redness), b* (yellowness), and IMF (intramuscular fat percentage). CM: surgically castrated

514 males; IM: immunocastrated males; EM: entire males; FE: females.

515

516

517 **SUPPLEMENTARY MATERIAL OF THE ARTICLE:**

518

519 **A matter of body weight and sex type: pig carcass chemical composition and pork quality**

520

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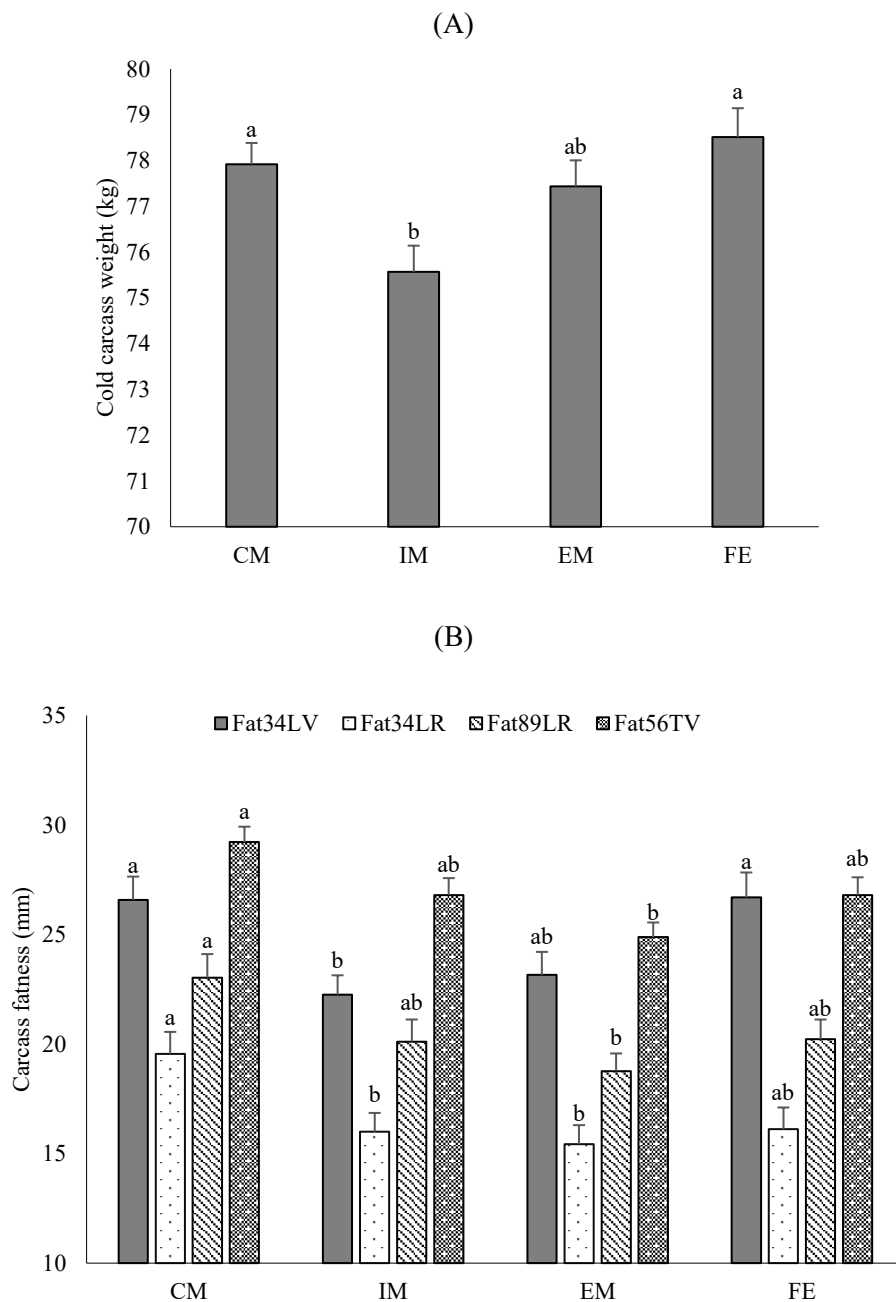
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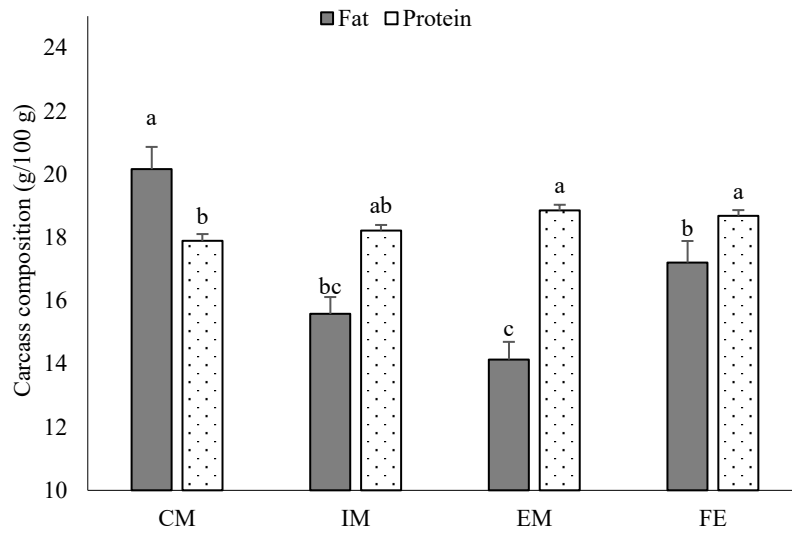
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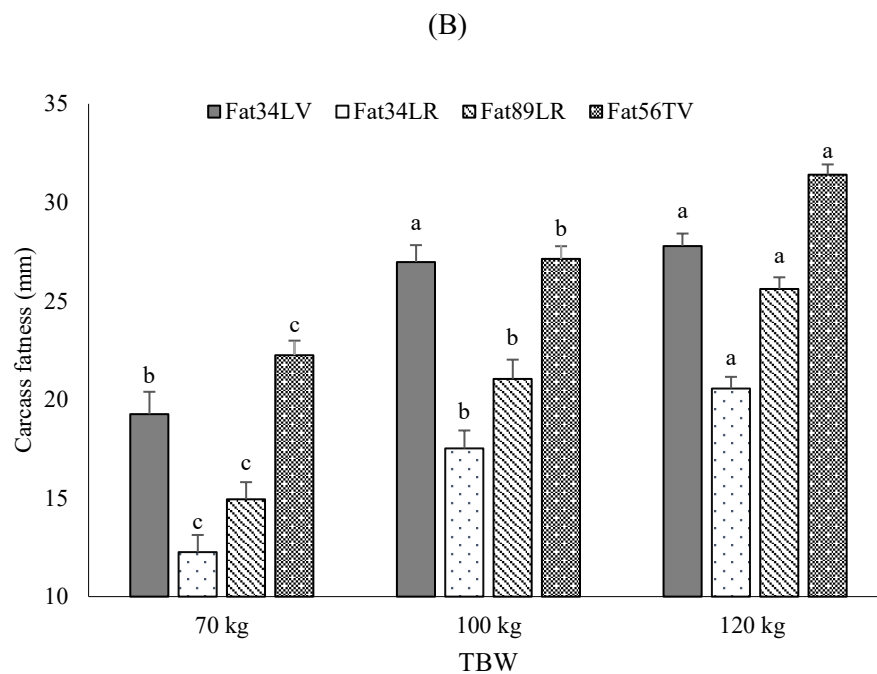
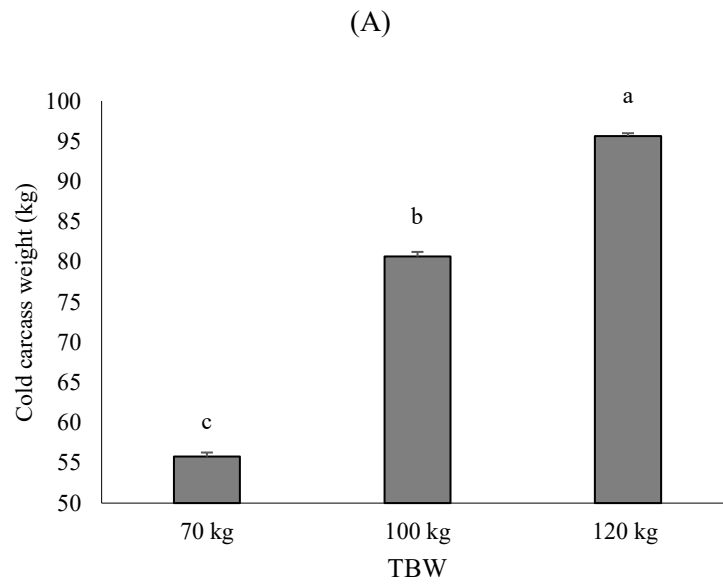


535 **Supplementary Figure 1.** (A) Cold carcass weight (kg) and (B) subcutaneous fat thickness (mm)
 536 (LSMEANS and SE) according to sex type averaged for all target body weights (70, 100, and 120
 537 kg). Different letters on error bars indicate significant differences between sex types (P -value \leq
 538 0.05). CM: surgically castrated males; IM: immunocastrated males; EM: entire males; FE:
 539 females. Fat34LV = fat thickness between the 3rd and 4th lumbar vertebrae; Fat34LR = fat
 540 thickness between the 3rd and 4th last ribs; Fat89LR = fat thickness between the 8th and 9th last
 541 ribs; Fat56TV = fat thickness between the 5th and 6th thoracic vertebrae.



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543 **Supplementary Figure 2.** Chemical composition of minced carcasses (g/100 g) (LSMEANS and
 544 SE) according to sex type averaged for all target body weights (70, 100, and 120 kg). Different
 545 letters on error bars indicate significant differences between sex types (P -value ≤ 0.05). CM:
 546 surgically castrated males; IM: immunocastrated males; EM: entire males; FE: females.



547 **Supplementary Figure 3.** (A) Cold carcass weight (kg) and (B) subcutaneous fat thickness (mm)
 548 (LSMEANS and SE) according to target body weight (TBW) averaged for all sex types
 549 (surgically castrated males, immunocastrated males, entire males, and females). Different letters
 550 on error bars indicate significant differences between TBW (P-value \leq 0.05). Fat34LV = fat
 551 thickness between the 3rd and 4th lumbar vertebrae; Fat34LR = fat thickness between the 3rd and
 552 4th last ribs; Fat89LR = fat thickness between the 8th and 9th last ribs; Fat56TV = fat thickness
 553 between the 5th and 6th thoracic vertebrae.

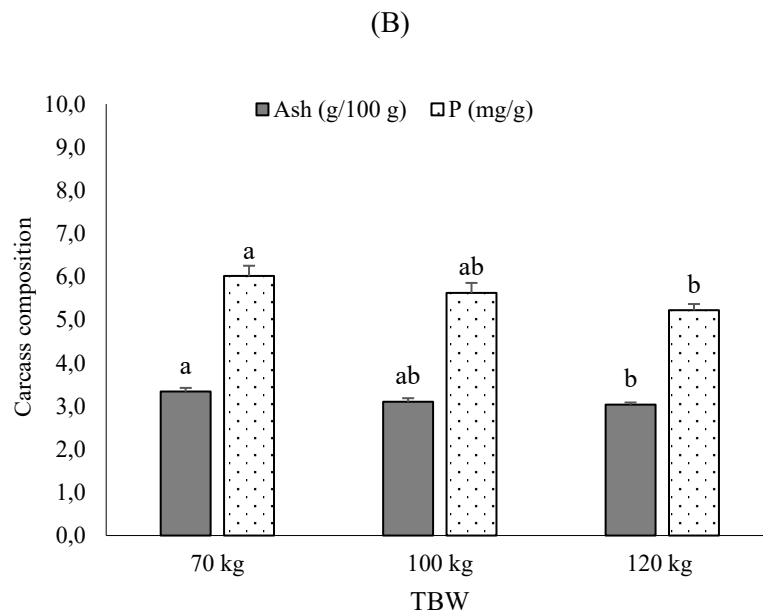
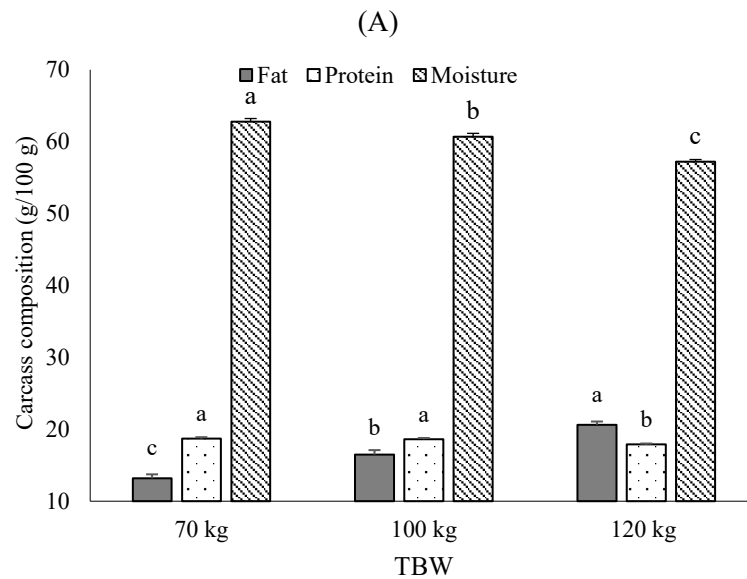
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559 **Supplementary Figure 4.** Chemical composition of minced carcasses (LSMEANS and SE)
 560 according to target body weight (TBW) averaged for all sex types (surgically castrated males,
 561 immunocastrated males, entire males, and females). Different letters on error bars indicate
 562 significant differences between TBW (P-value ≤ 0.05). (A) Fat, protein, and moisture content
 563 (g/100 g); (B) ash (g/100 g) and P content (mg/g).

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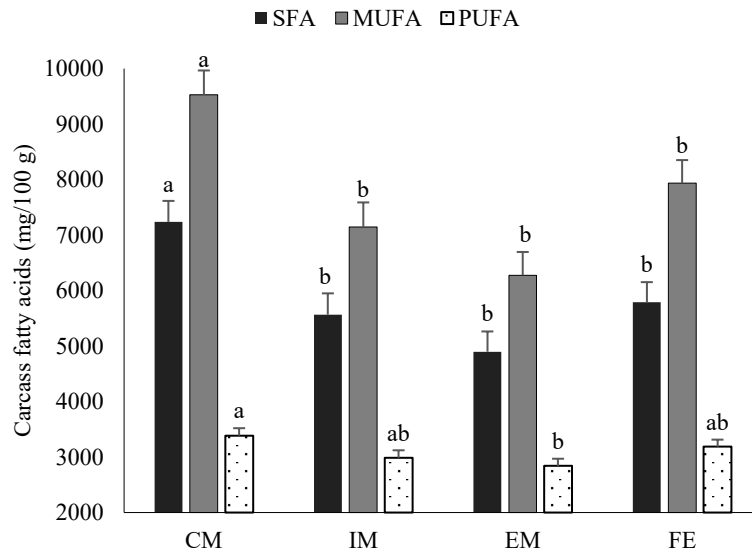
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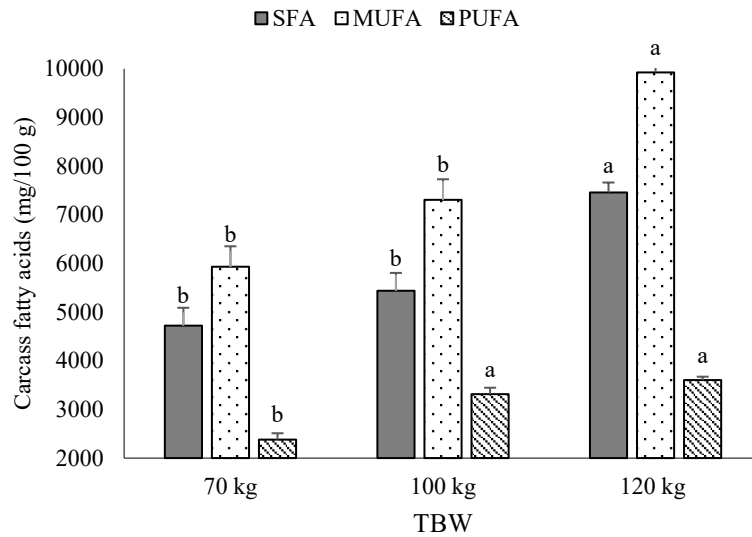
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573 **Supplementary Figure 5.** Saturated (SFA), monounsaturated (MUFA), and polyunsaturated
574 (PUFA) content of minced carcass (mg/100 g of minced carcass) (LSMEANS and SE) according
575 to sex type averaged for all target body weights (70, 100, and 120 kg). Different letters on error
576 bars indicate significant differences between sex types (P -value ≤ 0.05). CM: surgically castrated
577 males; IM: immunocastrated males; EM: entire males; FE: females.

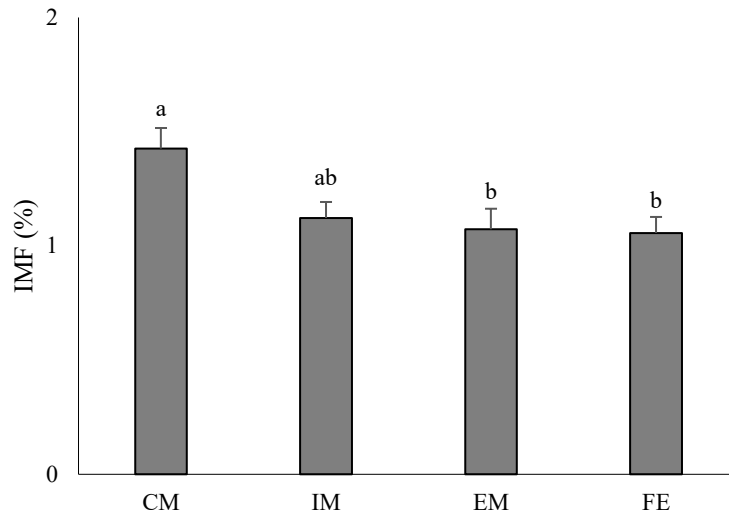
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580 **Supplementary Figure 6.** Saturated (SFA), monounsaturated (MUFA), and polyunsaturated
 581 (PUFA) fatty acid content of minced carcass (mg/100 g of minced carcass) (LSMEANS and SE)
 582 according to target body weight (TBW) averaged for all sex types (surgically castrated males,
 583 immunocastrated males, entire males, and females). Different letters on error bars indicate
 584 significant differences between TBW (P-value ≤ 0.05).

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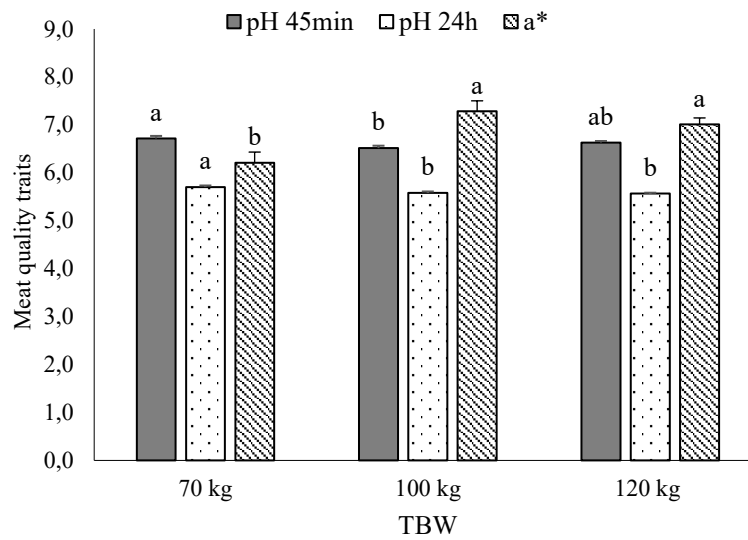


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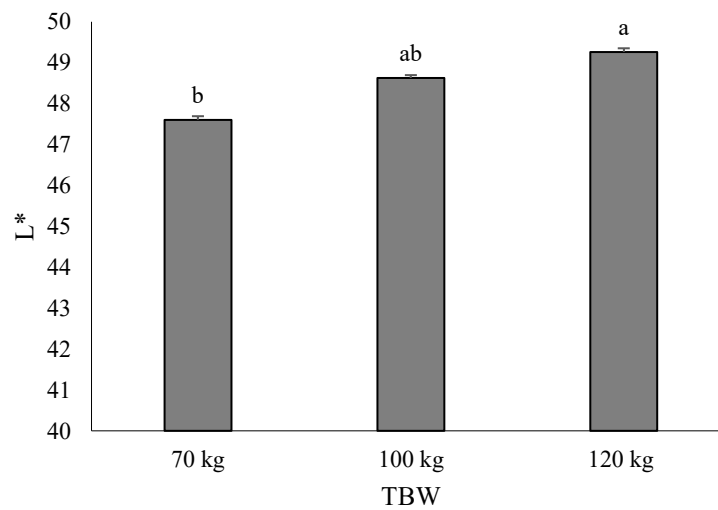
587 **Supplementary Figure 7.** Intramuscular fat percentage (IMF) of *Longissimus thoracis*
 588 (LSMEANS and SE) according to sex type averaged for all target body weights (70, 100, and 120
 589 kg). Different letters on error bars indicate significant differences between sex types (P-value ≤
 590 0.05). CM: surgically castrated males; IM: immunocastrated males; EM: entire males; FE:
 591 females.

Supplementary Figure 8.

(A)



(B)



592 **Supplementary Figure 8.** Meat quality traits of *Longissimus thoracis* according to target body
 593 weight (TBW) averaged for all sex types (surgically castrated males, immunocastrated males,
 594 entire males, and females). Different letters on error bars indicate significant differences between
 595 TBW (P-value ≤ 0.05). (A) pH value at 45 min, pH value at 24 h, and redness (a*); (B) luminosity
 596 (L*).

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