

Certain relationships between Animal Performance, Sensory Quality and Nutritional Quality can be generalized between various experiments on animal of similar types

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1	Certain relationships between Animal Performance, Sensory Quality and Nutritional
2	Quality can be generalized between various experiments on animal of similar types
3	
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15	
16	Abstract
17	In the beef sector, one of the major challenges is to early predict carcass and meat quality and to
18	jointly satisfy the multiple expectations of the various stakeholders. Thus, the objective of this
19	study was to determine if the relationships among carcass, nutritional and sensory qualities
20	established previously by Ellies-Oury et al. (2016) might be generalized to different type of
21	animals.
22	The Longissimus thoracis muscles of 32 young Charolais bulls were analyzed in terms of sensory
23	and nutritional quality (lipid content and fatty acid composition). These parameters of interest
24	were linked together and to animal performances by using a clustering of variables.

Longissimus thoracis sensory and nutritional qualities appear sometimes antagonistic. Indeed, some "positive" sensory descriptors (juiciness, overall appreciation, overall flavor and overall odor) are negatively related to PUFA proportions. PUFA proportions are positively associated with carcass weight but in the same time with rancid/fish flavors. Moreover, CLA and *trans* MUFA proportions are positively associated with the "negative" descriptors of greasy feel and residues. To finish, carcass weight and ADG are negatively associated with some "positive" sensory descriptors except tenderness.

32 It can be concluded from this work that these relationships, that were already established in33 previous works, are robust between experiments.

In order to highlight robust and generalizable relationships in different contexts, it is now appropriate to apply this method to a larger database containing different traits and various characteristics of breed, slaughter ages, animal types, fattening practices, ...

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38 Keywords

clustering of variables, animal performances, meat quality traits, fatty acid composition,nutritional value, prediction.

41

42 **1. Introduction**

The beef industry is made up of a set of links from farmers to consumers, through processors. Each of these intermediaries has specific expectations that are not necessarily consistent with each other. Among them, most of farmers have mainly expectations in terms of animal performance. In addition to the type of animal and its age, the carcass payment scale in Europe is mainly based on 3 parameters: weight, fattening state and conformation. Apart from a few specifications for which sensory and/or nutritional properties are specific expectations, otherwise, in most cases, these parameters are not taken into account in the remuneration of breeders(Monteils et al., 2017).

In cattle farming, as in most livestock farming, animal feed is the first item of expenditure (50% to 60% on average). It is therefore in the interests of farmers to select and breed efficient animals, that is to say, animals that are able to convert efficiently distributed feed into sales products (Ellies-Oury et al., 2020). This can have consequences on the nutritional and/or sensory properties of the meat from their carcasses.

Meat stakeholders main concern is the market structure and consumers' demands. The quality of carcasses (in terms of weight, composition or yields for example) is thus an important parameter for the meat sector, insofar as it determines the payment of the farmer, the remuneration of the intermediate link and the assurance of an optimized meat quality (i.e. satisfying the sometimes antagonistic interests of the various stakeholders of the chain ; Dockès et al., 2011).

In general, Europeans claim they want to favour food products that are good for their health in the coming years (Ellies-Oury et al., 2019; Hocquette et al., 2018). Meats have a relatively homogenous nutritional composition, at least for proteins. However, their content in lipids (ranging from 1-2% to around 15%; (Li, 2017; Normand et al., 2005)) and in saturated fatty acids (some of them being assumed to be detrimental to human health; (McAfee et al., 2010)) are quite variable. These heterogeneous compositions are thus likely to disturb the consumer.

The price of meat is the most important purchase criterion for 78% of the French, ahead of taste quality (46%) (Centre d'études et de Prospective, 2014). Ellies-Oury et al. (2019) specify in a recent study that 88% of the respondents would be interested in a system that would ensure a guaranteed level of meat tenderness and meat sensory quality at the time of purchase. Furthermore, 95% of these people would be willing to pay more for a cut of meat if such a system is implemented. Prediction models have previously attempted to establish response laws

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of muscle properties as a function of animal performances at farm or husbandry factors (Conanec 73 74 et al., 2019; Ellies-Oury et al., 2020; Ellies-Oury et al., 2019; Hocquette et al., 2017; Soulat et al., 75 2018, 2016). Other work has aimed to establish a model for predicting meat quality based on 76 carcass properties (Ellies-Oury et al., 2020; Ellies-Oury et al., 2019; Gagaoua et al., 2018). 77 However, in the light of these various studies, it appears that the response laws and prediction 78 models cannot always been scaled up through experiments. Indeed, for example, concerning 79 tenderness, Gagaoua et al. (2018) indicate that the best and interesting discriminators were 80 fattening duration and dry matter intake, whereas, according to Soulat et al. (2018), meat traits 81 were improved by the genetic of heifers' parents (*i.e.*, calving ease and early muscularity) and 82 when heifers were slaughtered older. The aim of this work was thus to determine whether the 83 relations highlighted between the different parameters of interest to the beef sector (animal performance, sensory quantity and nutritional quality) could be validated between 84 85 experiments. To do this, the method developed by Ellies-Oury et al. (2016) was applied in a 86 similar way to the data collected in the present experiment. Briefly, we used the *ClustOfVar* 87 approach on data established on animals of different breeds from those studied by Ellies-Oury et al. (2016). Developed to arrange variables into homogeneous clusters, this method allows 88 89 dimension reduction and variable selection (Chavent et al., 2011). Thus, this study should make it 90 possible to show whether the equations proposed by Ellies-Oury et al. (2016) are robust and 91 generalizable to different types of production. The ultimate objective of this work is, in the long 92 term, to predict meat qualities and performances based on different animal husbandry practices, 93 in order to manage these practices and, ultimately, to achieve quality and performance objectives 94 in line with demand.

95

96 2. Material and Methods

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Animal breeding and slaughtering procedures respected the French animal protection legislation,
including licensing of experimenters. The French Veterinary Services controlled and approved
the procedures at the slaughterhouse and at experimental facilities.

101

102 **2.1 Animal's management and slaughtering**

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All the experimental procedures performed in this study were approved by the Animal Ethics
Committee of INRA-CIRAD-IFREMER (APAFIS#1765-2015091516305 V3).

A total of 32 young Charolais bulls $(313 \pm 29.5 \text{ days old}, \text{mean live weight } 482 \pm 25.8 \text{ kg})$ were given individually for 6 months a basal diet consisting in 60% roughage (40% of corn silage and 20% of grass silage) and 40% of concentrate (Herbipole, INRAE, 2018). Daily feed intake and animal's body weight were recorded every 2 weeks. The average daily gain during growth (ADG during growth) was calculated from birth to weaning (estimated to 45 kg) but also during the growth and the finishing period. The ingested quantities were recorded every two weeks, allowing to estimate total and averaged ingested quantities of each component of the diet.

Animals were slaughtered in the experimental slaughterhouse of the INRAE Research Centre in which the slaughter rates are more constrained than in a commercial slaughterhouse. That's why animals were slaughtered in four times (at four different slaughter date) at the same body weight (736 \pm 39.5 kg). After 24 hours of food deprivation, animals were slaughtered in the experimental abattoir of the INRAE Research Centre, under standard conditions and in compliance with French welfare regulations. The carcasses were not electrically stimulated. They were chilled at 2°C in drying chambers during 24 hours, then stored at 4°C. A pH meter equipped with a glass

- electrode allowed to verify the ultimate pH 24h *post-mortem*. The aim was to have a pH in the *Longissimus thoracis* located between 5.7 and 5.9 between the 6th and 7th rib.
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123 **2.2 Animal performances**

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125 Average daily gain during the finishing period (ADG during the finishing period), finishing feed 126 conversion efficiency (ADG during the finishing period / dry matter) and finishing consumption 127 index (dry matter / ADG during the finishing period) were calculated using feed consumption 128 (expressed as dry matter or energy intake) during the finishing period and the live weight at the 129 beginning of the finishing period. Live weight and slaughter age were recorded at the 130 slaughterhouse. Carcass weight was then determined at slaughter. The removal and dissection of 131 the sixth rib joint allowed to assess its muscle, fat and bone proportions. According to Robelin & 132 Geay (1975) regression equations, the composition of the carcass (in content and in proportions) 133 was estimated, by using 1) the sixth rib muscle, fat and bone proportions and 2) the carcass 134 weight and measurement of the fatty deposits of the 5th quarter (Alberti et al., 2008). Carcass 135 yield was also determined.

- 136 The amounts in vitamin A and E in plasma were determined by HPLC-UV spectrophotometry137 adapted to bovine plasma (Scislowski et al., 2005).
- Total antioxidant status (SAO) in plasma was measured by a method based on the absorbance of
 the ABTS°+ cation [2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)] previously described
 by (Gobert et al., 2009).
- 141 Plasma Malondialdehyde (MDA) was determined by HPLC fluorescence as described earlier by
- 142 (Agarwal and Chase, 2002).

These parameters, evaluated on the blood of the animals, were considered to be relevant to animal performance, in particular because of the correlation that exists between efficiency and the antioxidant status of the animal as reported by Chauhan et al. (2016) in lambs and more generally because dietary antioxidants intake are known to affect growth (Catoni et al., 2008)

Thus, animal performances were characterized by 26 variables (Table 1). Means of each of the 26
variables studied in this experiment are given in Supplementary Data.

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150

151 **2.3 Muscle sampling**

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153 Samples (around 700 g) of the *Longissimus thoracis* (LT) muscle from the 10th thoracic rib were 154 collected 1-day *post mortem*. The muscle samples were conditioned and stored differently 155 according to the further analyses.

156 Samples for sensory evaluation (around 500 g) were cut into steaks, vacuum packed and kept at 157 4°C for ageing (14 days). Each sample was then frozen and stored at -20°C awaiting sensory 158 evaluation. A part of the remaining samples (50 g) was frozen in liquid nitrogen and kept at -159 80°C until analyses to determine enzyme activities, protein extraction and myosin heavy chain 160 (MyHC) quantification. Another part of the samples (150g) was cut into pieces (around 0.5 cm 161 cross-section), ground into fine and homogeneous powder in liquid nitrogen with a mixer mill 162 (Retch MM 301, Hann Germany) and stored at -80°C until analyses for collagen determination, 163 fatty acid composition and intramuscular fat content. Samples for one similar analysis were taken 164 at the same anatomical position from one animal to another.

165

166 **2.4. Muscular properties and sensory analysis**

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were measured according to procedure previously described by Dubost et al. (2013a).
Myofibrillar proteins from the LT muscles were extracted on ice as previously described by
Picard et al. (1994). The proteins were separated by SDS-PAGE electrophoresis according to
Picard et al. (2011). The proportions of the various myosin heavy chains (MyHC) were quantified
using densitometry with ImageQuant Software 5500 (Amersham Biosciences/GE Healthcare).

Total, insoluble and soluble collagen, cross-links (CLs) and proteoglycans (PG) of LT muscles

174 MyHC-IIb percentages were totalled with those of MyHC-IIx.

175 Sensory analyses were done at INRAE "Le Magneraud" station. After 14 days of ageing, each 176 sample was thawed at 4°C over 24 hours. The meat samples were cut into 1.5 thick steaks and 177 cooked under domestic grills until the temperature measured with a temperature probe reached 178 55°C (typical in France). Steaks were grilled between 2 aluminum sheets, until the end-point 179 temperature previously indicated in the geometric center of the steak was reached. After grilling, 180 each steak was cut into portions that were immediately presented to 12 panellists trained in beef 181 meat sensory analysis. Each sample was rated between 0 to 10 on an unstructured scale for the 182 following attributes: tenderness, juiciness, overall appreciation, overall odor, overall flavor, 183 rancid flavor, rancid odor, blood flavor, blood odor, fish flavor, fatty flavor, greasy feel, residues. 184 The score 0 represented a very low rating of the descriptor (extremely tough, very dry, ...) as 185 opposed to the score 10 which corresponds to a very high rating (extremely tender, extremely 186 juicy, ...).

187 Six sensory session were organized. At each of these sessions, the 12 panelists evaluated 5 or 6
188 samples randomly selected. Each sample was assessed by each panelist.

189 The expert panelists were trained in accordance with the ISO standards ISO/TC as described by190 Gagaoua et al. (2016). The sessions were carried out in a sensory analysis room equipped with

individual booths under artificial red light, in order to reduce the influence of the appearance of the samples. Each tasting booth was equipped with computer terminal linked to a fileserver running a sensory software (Fizz, version 2.20h; Biosystemes, Couternon, France) that facilitated the direct entry of assessor ratings. The variables composing sensory quality are reported in the table 2. Means of each of these variables studied in this experiment are given in Supplementary Data.

197

198 **2.5. Fatty acids composition**

199

200 Total lipids of LT muscle were extracted according to Folch et al. (1957) and quantified by 201 gravimetry. The FAs were extracted from total lipids and then converted into methyl esters by 202 transmethylation using borontrifluoride-methanol (14% solution) according the method of 203 (Bauchart et al., 2005). Fatty acid methyl ester analysis was performed by GLC using a Peri 2100 204 chromatography system (Perichrom, Saulx-les-Chartreux, France) fitted with a CP-Sil 88 glass 205 capillary column (Varian, Palo Alto, CA; length 100 m, dia. 0.25 mm) as previously described by 206 Gruffat et al. (2020). The carrier gas was H₂, and the oven and flame ionization detector 207 temperatures were as follows: oven temperature was programmed for 70°C for 30 s, 70 to 175°C 208 at a rate of 20°C/min, 175°C for 25 min, then 175 to 215°C at a rate of 10°C/min, and finally 209 215°C for 41 min; injector and detector temperatures were 235 and 250°C, respectively. Total 210 FAs were quantified using 19:0 as an internal standard. Their identification and the calculation of 211 the response coefficients were done using a C4-C24 quantitative mix (Supelco, Bellafonte, PA). 212 The variables composing nutritional quality are reported in the table 3. Means of each of these

213 variables studied in this experiment are given in Supplementary Data.

214

215 **2.6. Statistical analysis**

216

As previously indicated, in order to determine whether the associations observed between the different Parameters of Interest (PI: animal performance, sensory quantity and nutritional quality) can be scaled up through experiments, we applied the *ClustOfVar* (clustering of variables; (Chavent et al., 2011)) approach to the data collected in the present experiment for LT muscle (as this muscle was used in (Ellies-Oury et al., 2016) previous work).

222 Briefly, the proposed statistical methodology splits into 3 steps.

First, for each of the three PI, a hierarchical clustering of variables was applied to assess the links between the variables (within each PI) [step 1, figure 1]. The production of a tree structure within the variables allows to highlight hierarchical links between variables and to detect an ideal number of classes within the population (by defining a cut-off level). Clusters, grouping together the associated variables, were thus created. For each PI and each cluster, an Intermediate Score (IS) was calculated [step 1], the numerical IS summarizes all the numerical or categorical variables within the cluster.

230 More precisely, in the *ClustOfVar* method, the IS of a cluster is defined as the first principal 231 component obtained from the *PCAmix* (principal component analysis for mixed data) method 232 applied on the variables within the considered cluster. The underlying homogeneity criterion of 233 a cluster is defined as the sum of correlation ratios (for categorical variables) and squared 234 Pearson correlations (for numerical variables) to the synthetic numerical variable (the IS in the 235 proposed statistical methodology), summarizing "as good as possible" all the variables in the 236 cluster. Thus, a cluster of variables is defined as homogeneous when all the variables in the

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237 cluster are strongly linked to the corresponding numerical synthetic variable (the IS). Note that, 238 more generally, the numerical synthetic variables of the clusters can also be used for dimension 239 reduction or for recoding purpose. The IS scores reflect all the information contained in the 240 obtained clusters as long as each cluster is homogeneous. To determine the suitable number of 241 clusters for a given set of variables, a bootstrap approach has been also developed to evaluate 242 the stability of the partitions of variables and then provide the best partition (Chavent et al., 243 2011). In the following, to interpret the IS, only the variables within the cluster having a squared 244 correlation with the IS greater than 0.50 were used. Note that the variables selected may be 245 positively or negatively correlated with the IS.

Then, a second *ClustOfVar* analysis was done on all the previously created IS, allowing to build Global Indexes (GI) that were characterized by the IS's that are the best correlated to these GI (square correlation greater than 0.50) [step 2, figure 1].

Finally, in step 3 [step 2, figure 1], these GI were lastly compared between the two experiments in order to highlight robust relationships. That for, variable associations within each GI were compared. Stable pairings from one experiment to another (constituting equivalent clusters) could thus be identified. Conversely, it was also possible to highlight the variables for which the associations were unstable, i.e. the variables that changed cluster between the two experiments.

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3. RESULTS and DISCUSSION

3.1. Determination of linked variables in the present experiment

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The first result is that it was possible to constitute clusters of variables and thus build Intermediate Scores (IS). In the first step, 5 Intermediate Scores [denoted AP1* to AP5*] were retained concerning Animal Performances (figure 2) and respectively 4 and 5 IS concerning Sensorial [SQ1* to SQ4*] (figure 3) and Nutritional Quality [NQ1* to NQ5*] (figure 4). The increase of each of these IS led to either an increase or a decrease of the variables that compose the cluster, as indicated in table 4.

The clusters associated variables known to be correlated, especially because the number of clusters for each PI is relatively high compared to the total number of variables per PI: 5 vs. 26 for Animal Performances, 4 vs. 20 for Sensory Quality and 5 vs. 57 for Nutritional Quality. As for Ellies-Oury et al (2016), the synthetic indexes highlighted reflect widely accepted relationships such as:

²⁶⁹ - the positive link between food efficiency and ADG,

270 - the positive link between the different sensory parameters,

- the negative association between PUFA's proportions and the proportions of SFA and MUFA,
272

In the second step, it was possible to build Global Indexes (GI) by making clusters within Intermediate Scores. Four GI were highlighted [GI1* to GI4*] (table 5; figure 5). With the exception of GI4*, which associated only ISs related to animal performance [AP2*, AP3* and AP5*], all other GIs associated ISs related to different PI:

<u>- GI1* associated positively NQ3*, SQ1* and SQ4*.</u> Thus, in this GI, the proportions of
 CLA and *trans* MUFA were positively associated with the sensorial descriptors of fatty flavor,
 rancid flavor, greasy feel and residues, but also with MyHCIIa isoforms and collagen content
 (total, insoluble and CLs contents). All these parameters were negatively linked to tenderness.

<u>- GI2* associated positively AP1* and negatively NQ1*, NQ2*, NQ4* and SQ3*.</u> The
 SQ2* Intermediate Score was also negatively associated in this GI, but as the square correlation
 was lower than 0.50, this IS was removed for the GI2* interpretation. In this GI, the proportions

of PUFA were positively associated with live and carcass weights but also with rancid and fish flavors. All these variables were negatively linked to the amounts of lipids, the amounts and proportions of SFA and MUFA, but also to the positive descriptors of juiciness, overall flavor, overall odor and overall appreciation.

<u>- GI3* associates positively AP4* and NQ5*.</u> According to the interrelations identified in
 this GI, the amounts of PUFA were positively associated with slaughter age and ADG between
 birth and weaning.

291 To finish with, the GI4* associated ingested quantities, feed efficiency, carcass yield and muscle
292 development, finishing growth rate and MDA content.

293

294 As noted earlier, the *ClustOfVar* method allows to group together variables that share similar 295 information and that are thus highly correlated to each other. The advantage of this method is that, 296 in the same analysis, different types of variables (contents, proportions, ratios, ...) can be grouped 297 together, without any problem of collinearity. This specificity of the clustering of variables is 298 clearly useful in our case, in that some of the different variables used are in part redundant with 299 each other. On the other hand, in the method used here, two successive clustering of variables 300 were carried out (steps 1 and 2) in order to compare our results with those established by Ellies-301 Oury et al. (2016). This succession of clustering is at the origin of a loss of information that is 302 likely to limit the identification of relationships between the studied variables all the more as, as 303 previously indicated, only the variables having a square correlation with the IS greater than 0.50 304 were used.

The variable clustering approach aims to maximize a homogeneity criterion among the highly correlated variables. However, from one experiment to another one, the available variables might be different and the homogeneity criterion might thus be different. To finish with, what might also be underlined, the stability of partitions of variables was evaluated with bootstrap approach (Chavent et al., 2011). However, other methods might have been developed to determine the suitable number of clusters of variables, and might have conducted to a number of clusters different, and thus to different associations between variables. Nevertheless, the evaluation of the associated variables under other conditions of determination of the number of clusters makes it possible to highlight that the general tendencies remain unchanged whatever the number of clusters finally formed.

315

In the light of the associations of variables found in this paper, it can be shown that the main conclusions previously established by Ellies-Oury et al. (2016) on the LT muscle of Angus, Blondes d'Aquitaine and Limousin bulls were also true in this new experiment on the same muscle collected on Charolais bulls.

320

321 For example, we established that juiciness and the descriptors of overall appreciation, overall 322 flavor and overall odor were positively related to meat lipid content but negatively related to the 323 proportions of PUFA and EPA. Moreover, it appears that the positive sensory descriptors of 324 juiciness as well as the overall odor, flavor and appreciation descriptors were negatively 325 associated to carcass weight and ADG during growth. It can be hypothesized that this negative 326 association is related to lower fat development of animals with a high ADG during growth, as 327 already established by Ellies Oury et al. (2016). Indeed, during growth, most of nutrients are used 328 for muscle development instead of fat development and thus, intramuscular fat (IMF) is deposited 329 at a lower rate than muscle proteins. On the contrary, IMF is deposited at a greater rate than muscle proteins during finishing period, since less nutrients are used for muscle growth (Pethick 330 331 et al., 2004).

332 Positive sensory descriptors of juiciness, overall appreciation, overall flavor and overall odor 333 were also negatively associated with nutritional value of meat. As described above, increasing 334 lipid content in meat led in most publications to an increase in sensorial descriptors of tenderness, 335 juiciness, flavor and overall acceptability scores in trained jury but also in consumer sensory 336 panels (Garmyn et al., 2011; Hocquette et al., 2010; Jeremiah et al., 2003; Lorenzen et al., 2003; 337 May et al., 1992; Mottram, 1998; Neely et al., 1998; O'Quinn et al., 2012; Savell et al., 1987; 338 Smith et al., 1985). However, even if fat and fatty acids contribute to a large extent to meat 339 sensory quality, they also contribute to the nutritional value of meat (Wood et al., 2008; Scollan 340 et al., 2001, 2014; Wyness et al., 2011). Indeed, for human, the relationships between dietary fat, 341 irrespective of the food source, with the incidence of various lifestyle diseases including 342 cardiovascular diseases is well established and several health agencies have proposed specific 343 guidelines. Some fatty acids are also known to affect human health in several ways, especially 344 some SFAs, such as C16:0, that is known for their harmful properties on human health 345 (Muchenje et al., 2009). In the case of beef, the fatty acids present in the adipocytes being mostly 346 SFAs and MUFAs (Wood et al., 2008), fatter animals have higher SFA proportions in their 347 muscles and, thus, deleterious fatty acids.

348 However, juiciness, flavor/odor and overall appreciations are positively related to amounts and 349 proportions of C18:1, confirming by the same way, that the fatter the animals are, the higher the 350 proportion of SFA and MUFA (and therefore of C18:1 which is the major fatty acid in MUFA) in 351 the meat, the juicier the meat. These results confirm the conclusions of various studies (Dryden 352 and Maechello, 1970; Garmyn et al., 2011; Melton et al., 1982; Westerling and Hedrick, 1979), 353 that establish a positive correlation between beef palatability and C18:1. Moreover, this FA is 354 known to be a "neutral" FA without any deleterious or beneficial impact on human health 355 (Pereira and Vicente, 2013).

In addition, as previously indicated by Ellies Oury et al. (2016), juiciness and the descriptors of 356 357 overall appreciation, overall flavor and overall odor were positively related to meat lipid content 358 but negatively related to the proportions of PUFA and EPA. This result is logical since, as 359 explained above, the increase in lipids in meat leads to an increase in the proportions of SFA and MUFA and thus a decrease in the proportion of PUFA. PUFAs are widely recognized for their 360 361 beneficial properties for human health, including EPA and DHA which can profoundly influence human health (Vahmani et al., 2015). Thus, once again, juiciness and the descriptors of overall 362 363 appreciation, overall flavor and overall odor are negatively correlated with the health value of 364 meat. Surprisingly, the tenderness is negatively related to the proportions of CLA and CLA 365 precursors. These FA are known to reduce body fat, cardiovascular diseases and cancer, and to 366 modulate immune and inflammatory responses (Dilzer and Park, 2012; Whigham et al., 2000). 367 This negative relationship is surprising since CLAs are mainly stored in triglycerides (that are 368 storage lipids that increase with body fat). According to the literature, tenderness might also be 369 negatively correlated to PUFA proportions, whereas its correlation with MUFA appears positive 370 (Garmyn et al., 2011). In conclusion, it seems complicated to jointly obtain a meat with 371 optimized sensory and nutritional properties. There is a need to understand the relationship 372 between palatability, fatty acid composition and fat content, in order to ensure subsequently that 373 tenderness, flavor, and juiciness are not compromised when selecting cattle with enhanced 374 nutritional composition.

In the present work, n-3 and n-6 PUFA proportions were positively linked (and negatively linked to n-3 and n-6 PUFA amounts, these amounts being positively linked together). These conclusions are contrary to those of the literature as in some studies, increasing n-3 PUFA proportions matched with decreasing n-6 PUFA proportions, showing competition between these fatty acid families for the same set of elongation and desaturation enzymes (Lorenz et al., 2002;

Nuernberg et al., 2005). Working on various bovine breeds, (Ellies Oury et al., 2016) indicated 380 381 that increasing n-3 PUFA proportions leads to an increase of n-6 PUFA proportions (n-3 PUFA 382 amounts are also positively correlated with n 6 PUFA amounts), but this correlation was found to 383 be breed dependent. In fact, in the literature, most studies compared diets rich in n 3 PUFA to diets rich in n 6 PUFA. It is then normal that an increase in one family of fatty acids is associated 384 385 with a decrease in the other. In the present experiment, the animals were all fed the same diet (40% of corn silage, 20% of grass silage and 40% of concentrates) which provided both n 6 (corn 386 387 and concentrates) and n-3 (grass silage) PUFA. Thus, the present results might stem from an 388 animal diet impact, as it is widely accepted that cattle feed (enriched or not with certain PUFAs and in particular n-3 PUFA) might have a significant impact on the FA composition of the meat. 389

390 If (Ellies-Oury et al., 2016) has previously revealed a positive relation between collagen content 391 and lipid content of beef, a link might here be established between collagen and cross link 392 contents and fat linked sensory descriptors such as fat flavor and greasy feel. All of these 393 variables are also positively associated with the proportions of CLA and CLA precursors.

Collagen composition (total and insoluble collagen, CLs) was positively associated with residues and negatively with tenderness. This conclusion, which is the opposite of that of (Ellies-Oury et al., 2016), is however consistent with the results previously established in the literature, which associated the increase in collagen content with an increase in meat hardness (Destefanis et al., 2000; Dransfield et al., 2003; Dubost et al., 2013b; Lepetit, 2007; McCormick, 1999; Nishimura et al., 2009; Renand et al., 2001; Rhee et al., 2004; Torrescano et al., 2003).

400 No relationship was found between the proportions of the different myosin isoforms and the
401 sensory quality of the meat, contrary to the results of the literature (Chriki et al., 2013; Crouse et
402 al., 1991; Ellies-Oury et al., 2016; Jurie et al., 2007; Maltin et al., 1998; Therkildsen et al., 2002;
403 Zamora et al., 1996), who established a positive association between oxidative metabolism and

404 sensory quality. Nevertheless, the relationship appears controversial as other authors, such as 405 (Picard et al., 2014), that have found a negative correlation between these same variables, 406 depending on the muscle type and/or the breed of the animals. Thus, even though several studies 407 reviewed by Guillemin et al. (2009) have previously considered that fiber metabolic and enzyme 408 activities are notably implicated in meat quality traits and especially in tenderness, no significant 409 interrelation are observed in the present work.

410 To finish, we logically establish that carcass butcher value (carcass yield and muscle 411 development in the carcass) is associated with lower carcass fatness but positively with 412 peroxidation intensity. As indicated above, the leaner the meat, the more PUFA proportions it 413 contains. However, it is well known that the PUFAs present in beef are sensitive to oxidation 414 especially as the level of antioxidants such as vitamins E and A is low (Durand et al., 2005). Thus, 415 it is not surprising that carcass butcher value is positively associated with MDA levels, that is an 416 index of peroxidation intensity and negatively associated with the total antioxidant status and 417 vitamin A and E levels of meat, that are indicators of the protection level of PUFA against 418 peroxidation (Durand et al., 2005). It would therefore be relevant to introduce antioxidants in the 419 diet of animals with a high carcass butcher value, in order to maintain the nutritional value of 420 their meat.

421

422 **4. CONCLUSION**

The objective of this work was to determine whether the relations highlighted between the different parameters of interest to the industry (animal performance, sensory quantity and nutritional quality) could be validated between experiments. In the present work, we used the *ClustOfVar* package to validate, with data from a new experiment, the relationships previously highlighted in the literature (Ellies-Oury et al., 2016). In the light of these associations of

18

428 variables, it can be shown that the main conclusions previously established on the LT muscles (of 429 Angus, Blonde d'Aquitaine and Limousine cattle), were also found in this new experiment on the 430 same muscle but on a different breed (Charolais). Indeed, the generalization of the equations 431 established between the Parameters of Interest can help to determine the compromises that it will be necessary to make in order to meet the expectations of the different links of the sector. It will 432 433 thus be possible to propose "realistic" specifications adapted to the different breeders / operators / 434 consumers. However, the generalizations have only been shown to apply for the Longissimus 435 thoracis from young bulls finished under feedlot conditions and slaughtered when they achieved 436 a set weight.

437

In order to widely generalize the established relationships, it is now appropriate to apply this
method to a database containing different experiments with various characteristics. It will thus be
possible to highlight robust and generalizable relationships in different contexts (different breeds,
types of animals, ...).

442 This method was carried out until now on the LT muscle, which is commonly considered as the 443 reference muscle. However, in the light of various studies, it appears that the response laws and 444 prediction models cannot always been scaled up through muscles within the same experiment. 445 Thus, now that it has been proven that the relationships between the Parameters of Interest 446 (animal performance, sensory quality, nutritional quality) can be transposed from one experiment 447 to another on the same muscle, it is necessary to determine whether the proximities established 448 between the three Parameters of Interest could be extrapolated from one muscle to another within 449 the same carcass. It would then be possible to predict the properties of all the muscles of the 450 carcass by knowing the properties of one of them. Such predictive equations of the nutritional 451 and sensory properties of the various muscles of the carcass would be of real interest to the

- 452 industry. Thus, in the long run, it should be possible to integrate these equations into a
 453 mathematical model for the management of animals in order to obtain meats whose properties
 454 will perfectly meet the expectations of the different markets / market niches.
- 455

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459

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- 471

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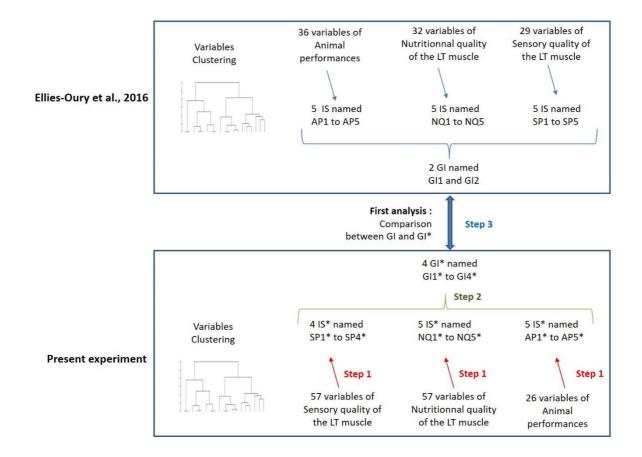


Figure 1: principle of the method developed by Ellies-Oury et al. (2016) and in the present experiment

IS: intermediate score – GI: global index – LT: *Longissimus thoracis* – AP: animal performance – NQ: nutritional quality – SQ: sensory quality

Cluster Dendrogram

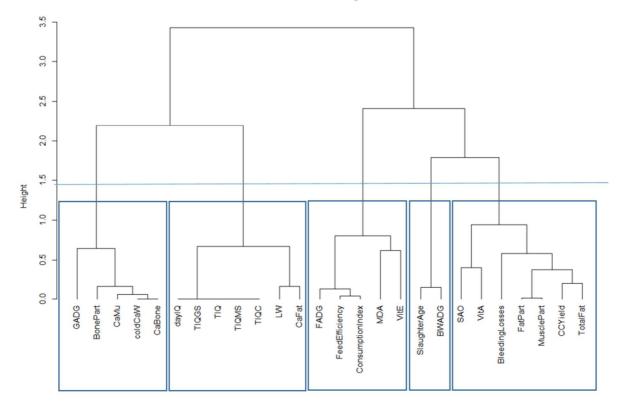


Figure 2: Intermediate Scores (n=5) retained concerning Animal Performances

Cluster Dendrogram

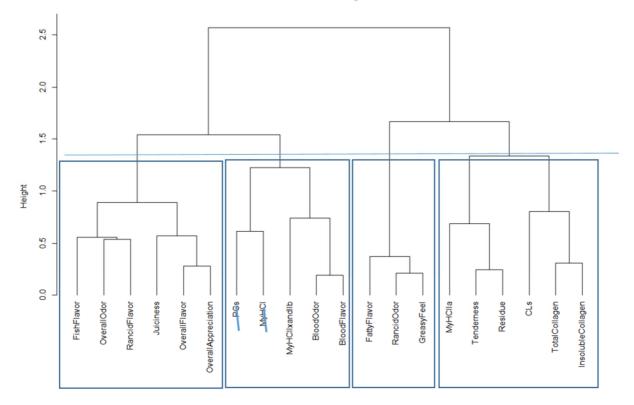


Figure 3: Intermediate Scores (n=4) retained concerning Sensorial Quality The 2 crossed-out variables (PGs, MyHCI) correspond to variables poorly represented in the cluster (and not analyzed in the following).

Cluster Dendrogram

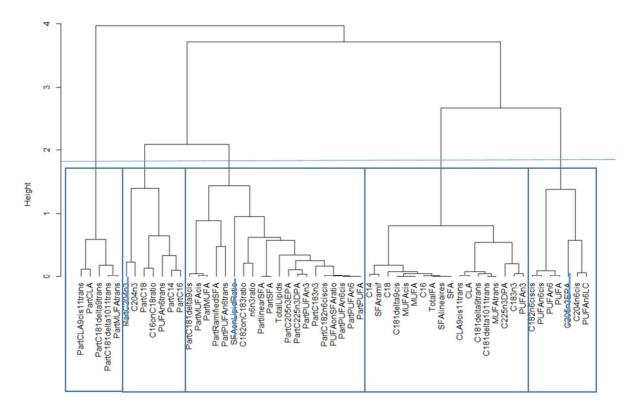


Figure 4: Intermediate Scores (n=5) retained concerning Nutritional Quality The 3 crossed-out variables (%C20:4n3; SFA/Lipid ratio; C20:5n3EPA) correspond to variables poorly represented in the cluster (and not analyzed in the following)

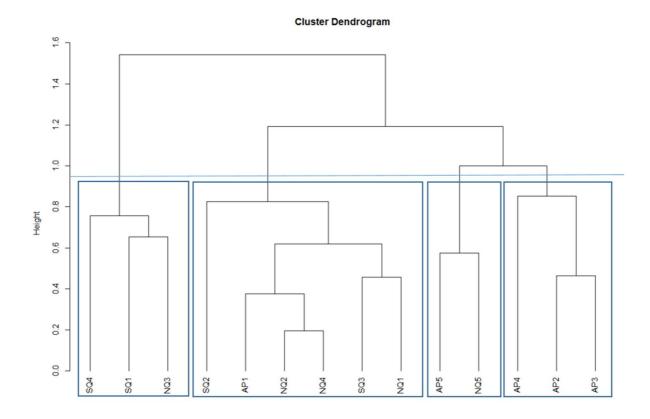


Figure 5: Global Indexes (GI) built by making clusters within Intermediate Scores 5 Intermediate Scores [denoted AP1* to AP5*] concerning Animal Performances 4 IS concerning Sensorial Quality [SQ1* to SQ4*] 5 IS concerning Nutritional Quality [NQ1* to NQ5*]

Table 1: list of the 26 variables of animal performances analyzed on the 32 young Charolais bulls

Variables

Age at slaughter; Total ingested quantities in grass silage (TIQGS), maize silage (TIQMS),

concentrate (TIQC); Total ingested quantities (TIQ); Ingested quantities per day (dayIQ);

Live weight (LW); Carcass weight (CaW);

Carcass Yield (CCYield = CaW / LW); Bleeding losses; Total fat weight (Total Fat);

Carcass fat weight (CaFat); Carcass proportion of fat (FatPart);

Carcass bone weight (CaBone); Carcass proportion of bone (BonePart);

Carcass muscle weight (CaMuscle); Carcass proportion of muscle (MusclePart);

Average Daily Gain (ADG) between birth and weaning (BWADG), during growth

(GADG), and during finishing period (FADG)

Feed conversion Efficiency (ADG / total dry matter intake); Consumption index (total dry

matter intake / ADG)

Amount in A and E vitamins;

MDA; SAO

Table 2: list of the 20 variables of sensory quality analyzed on the 32 young Charolais bulls

Variables

Tenderness; Juiciness; Residue; Overall appreciation; Overall odor; Overall Flavor; Fish

flavor; Blood odor; Blood flavor; Rancid odor; Rancid flavor; Fatty flavor; Greasy feel;

Total collagen; Insoluble collagen; CLs; PGs;

MyHC I; MyHC IIa ; MyHC IIx+b;

Table 3: list of the 57 variables of nutritional quality analyzed on the 32 young Charolais bulls

Variables **Fatty acid composition expressed in content (g or mg / 100 g of muscle):** Total Lipids; Total Fatty Acids (FA); Saturated FA (SFA); Monounsaturated FA (MUFA); Polyunsaturated FA (PUFA); C14:0; C16:0; C18:0; ramified SFA; linear SFA; C18:1 n-9 cis; *trans* C18:1 10/11; *trans* C18:1 n-9; *cis* MUFA; *trans* MUFA; C18:2 n-6; *cis* C20:4 n-6; PUFA n-6; C18:3 n-3; DPA; EPA; PUFA n-3; long chain PUFA n-3; Total Conjugated Linoleic Acids (CLA); CLA 9*cis* 11*trans*; *trans* PUFA; *cis* PUFA **Fatty acid composition expressed in proportions (% of total FA):** Total Fatty Acids; Saturated FA; Monounsaturated FA; Polyunsaturated FA; C14:0; C16:0; C18:0; ramified SFA; linear SFA; C18:1 n-9 cis; C18:1 9*trans*; C18:1 10/11*trans*; *cis* MUFA; *trans* MUFA; C18:2 n-6; C20:4 n-6; PUFA n-6; *trans* PUFA n-6; *cis* PUFA n-6; C18:3 n-3; EPA; DPA; PUFA n-3; Total Conjugated Linoleic Acids; CLA 9*cis* 11*trans* **Ratios :** C16:0 / C18:0; PUFA / SFA; PUFA n-6 / PUFA n-3 ; C18:2 n-6 / C18:3 n-3

Table 4: Composition of each of the Intermediate Scores obtained for each parameter of interest (animal performances, sensorial quality and nutritional quality) in both experiment

	Ellies-Oury et al., 2016	Present experiment
Animal Performances (AP)	AP1 Fat development in the carcass and in the 5 th quarter, but also total fat and trim fat	AP1* Carcass Weight, Amount of muscles/bone in the carcass, ADG during growth Negatively linked to Part of Bone in the carcass
	AP2 Empty body weight and bone development	AP2* Bleeding losses, Carcass yield, Part of muscle in the carcass, Negatively linked to Fat development in the carcass, amount in A vitamins, Part of fat in the carcass, SAO
	AP3 Feed conversion efficiency, ADG during finishing period	AP3* Feed conversion efficiency, ADG during finishing period, MDA Negatively linked to Consumption index, amount in E vitamins
An	AP4 Viscera weight and proportions	AP4* Live weight, Amount of fat in the carcass, ingested quantities (total, per day, GS, MS, C)
	AP5 Butcher value (compactness, thigh thickness, muscle development, yields, carcass weight)	AP5* Age and ADG between birth and weaning
	SQ1 Meat quality traits of steaks cooked at 55°C (higher tenderness and lower residue content) and/or 74°C (higher tenderness and juiciness and lower residue content)	SQ1* MyHCIIA, Residues, collagen amounts (total, insoluble) and CLs Negatively linked to Tenderness
Sensorial Quality (SQ)	SQ2 Whiteness (L*) Negatively linked to yellowness (b*)	SQ2* MyHC IIX and IIB, blood odor and flavor
orial Qua	SQ3 Oxidative metabolism (ICDH and COX activities and %MyHC I)	SQ3* Juiciness and Overall flavor/odor/appreciation Negatively linked to Rancid and fish flavors
Sens	SQ4 Collagen content and insolubility, lipid content Negatively linked to glycolytic fibres proportions (MyHC IIx+IIb)	SQ4* Fatty flavor, rancid odor and greasy feel
	SQ5 Protein content	
	NQ1 n-6/n-3 ratios (n-6/n-3 and C18:2 n-6 / C18: 3n-3)	NQ1* C16:0/C18:0, % C14:0, % C16:0, <i>trans</i> PUFA amount Negatively linked to % C18:0, % C20:4 n-3
Nutritional Quality (NQ)	NQ2 Total lipids, Amounts of SFA, CLA and MUFA	NQ2* Lipids, % SFA, % linear SFA, % MUFA, % <i>cis</i> MUFA, % <i>cis</i> C18:1D9 Negatively linked to C18:2 n-6/C18:3 n-3, n6/n3, % ramified SFA, PUFA/SFA % EPA, % DPA, % n-3 PUFA, % C18:2 n-6 <i>cis cis</i> , % C18:3 n-3, % <i>trans</i> n-6 PUFA, % <i>cis</i> n-6 PUFA, % n-6
utritional	NQ3 % MUFA <i>trans</i>	PUFA, %PUFA NQ3* % CLA, % CLA 9 <i>cis</i> 11 <i>trans</i> , % C18:1 9 <i>trans</i> , % C18:1 10-11 <i>trans</i> , % <i>trans</i> MUFA
Ň	NQ4 % PUFA, % n-6 PUFA, % n-3 PUFA Negatively linked to %SFA and %MUFA	NQ4* Amounts of FA, C14:0, C16:0, C18:0, ramified SFA, linear SFA, SFA, <i>cis</i> MUFA, MUFA, C18:1n-9 <i>cis</i> , CLA 9cis 11 <i>trans</i> , CLA, C18:1 9 <i>trans</i> , C18:1 10-11 <i>trans</i> , <i>trans</i> MUFA, C22:5 n-3 DPA, C18:3 n-3, n-3 PUFA
	NQ5	NQ5*

Amounts of PUFA, n-3 PUFA and n-6 PUFA	Amounts of PUFA, n-6 PUFA, long chain n-6 PUFA, cis
	n-6 PUFA, C18:2 n-6 cis cis, C20:4 n-6 cis

Table 5: Composition of each Global Index established by a clustering of the previous IS in both experiment

Ellies-Oury et al., 2016	Present experiment
GI1: + AP1 + NQ1 - SQ1 Butcher value (compactness, thigh thickness, muscle development, yields, carcass weight), % PUFA, % PUFA n- 3, % PUFA n-6, Glycolytic fibres, Residues Negatively linked to Fat development, Non-productive needs, Lipid, SFA, MUFA, PUFA, n-3 PUFA, n-6 PUFA, % SFA, % C14:0, % C16:0, % MUFA, Tenderness (55°C and 74°C), Juiciness (74°C), Oxidative metabolism, Collagen amounts (total, insoluble)	GI 1*: + NQ3* + SQ1* + SQ4* % CLA, % CLA 9 <i>cis</i> 11 <i>trans</i> , % C18 :1 9 <i>trans</i> , % C18 :1 10-11 <i>trans</i> , % <i>trans</i> MUFA MyHCIIA, Residues, collagen amounts (total, insoluble) and CLs Fatty flavor, rancid odor and greasy feel Negatively linked to Tenderness
GI2 = + NQ2 + SQ2 % MUFA <i>trans</i> , Yellowness (b*), Protein content Negatively linked to Whiteness (L*), n-6 / n-3, C18:2 n-6/C18:3 n-3	GI 2 *: + AP1* – NQ2* – NQ4* – NQ1* – SQ3* (-SQ2*) Carcass weight, amount of muscles/bone in the carcass, ADG during growth % C18:0, % C20:4 n-3, % EPA, % DPA, %, n-3 PUFA, % C18:2 n-6 <i>cis cis</i> , % n-6 PUFA, % PUFA, % C18:3 n-3, % ramified SFA, % <i>cis</i> n-6 PUFA, % <i>trans</i> n-6 PUFA, C18:2 n-6/C18:3 n-3, n-6/n-3, PUFA/SFA, Rancid and fish flavors Negatively linked to C16:0/C18:0, Lipids % SFA, % C14:0, % C16:0, % linear SFA, % MUFA, % <i>cis</i> MUFA, % C18:1 n-9 <i>cis</i> ; Amount of FA, C14:0, C16:0, C18:0, ramified SFA, linear SFA, SFA, <i>cis</i> MUFA, MUFA, C18:1 n-9 <i>cis</i> , CLA 9 <i>cis</i> 11 <i>trans</i> , CLA, C18:1 9 <i>trans</i> , C18:1 10-11 <i>trans</i> , <i>trans</i> MUFA, C22:5 n-3, C18:3 n-3, n-3 PUFA, <i>trans</i> PUFA, Juiciness and Overall flavor/odor/appreciation, Part of Bone in the carcass GI 3*: + NQ5* + AP4* Amount of PUFA, n-6 PUFA, long chain n-6 PUFA, <i>cis</i> n- 6 PUFA, C18:2 n-6 <i>cis cis</i> , C20:4 n-6 <i>cis</i> Age and ADG between birth and weaning GI 4*: + AP3* + AP2* + AP5* Live weight, amount of fat in the carcass, Ingested quantities, Bleeding losses, Carcass yield, Part of muscle in the carcass, Feed conversion efficiency, ADG during finishing period, MDA Negatively linked to Fat development in the carcass, Part of fat in the carcass, Consumption index, Amount in A and E vitamins, SAO