

# Ability of physico-chemical measurements to discriminate rabbit meat from three different productive processes

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## Abstract

**BACKGROUND:** To fulfil consumers' requirements for food traceability, it is necessary to have effective tools to differentiate food products according to their origin. The aim of the study was to identify a limited number of physico-chemical measurements that could differentiate rabbit meat from three different rearing systems: standard production system or a high quality norm system or a very low growth breeding system.

**RESULTS:** The stepwise linear discriminant analysis (LDA) provided 14 physico-chemical variables, then combined into two discriminant factors. Most of them ( $n = 8$ ) were related to bone traits, and especially ( $n = 5$ ) to mechanical femur assessments. Mechanical characteristics of meat were also relevant in this analysis. Decision tree analysis (DTA) selected two variables only (femur stiffness, and ratio of femur weight to chilled carcass weight) to discriminate the three groups. A total of 96% and 90% of rabbits were correctly assigned to their original group according to LDA and DTA, respectively.

**CONCLUSION:** This work demonstrated that simple physico-chemical traits recorded in carcasses and meat were efficient to discriminate rabbits from three different rearing systems using LDA or DTA procedures. These systems could have further implications for future traceability of breeding origin.

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**Keywords:** rabbit; meat quality; rearing system; discriminant analysis; decision tree

## INTRODUCTION

Forty-seven percent of world rabbit production arises from the European Union, where Italy, Spain and France are the top rabbit-meat producing countries (77% of the European Union production<sup>1</sup>). In this area, rabbit meat is considered as a highly nutritious, low-fat and low-cholesterol meat. Nevertheless, for all meats,<sup>2</sup> including rabbit, consumer perception of meat quality has been badly affected recently by various health crises, resulting in a decreased consumption of meat in Europe. To counteract this trend, new labelled production systems have been developed in which rabbits are reared under carefully specified conditions, in order to meet specific consumer requirements, i.e. information on origin, rearing conditions and guarantee on animal welfare, as well as to propose rabbit meat with high sensory qualities. In France, for example, product conformity certifications of rabbit production represented 15% of slaughtered rabbits in 2003<sup>3</sup> and a Label Rouge production exists as a niche

market. The main attributes that attract consumers to purchase rabbit Label Rouge production, despite the high price, is the guarantee of superior sensory qualities which arise from rearing conditions. In French Label Rouge production, the use of slow-growing genotype, low-energy feedstuff, and low stocking density resulting in a higher age at slaughter (not less than 91 days) compared with standard production (less than 70 days) are meant to improve sensory meat quality traits. However, reported malpractices have caused the public to spurn the product and have resulted in increased requirements for traceability. This means having effective tools to objectively measure additional qualities. The ability of physico-chemical measurements to differentiate food products according to their origin has previously been proved (e.g. in trout,<sup>4</sup> wine,<sup>5</sup> and honey<sup>6</sup>). In rabbit, previous studies<sup>7,8</sup> have estimated the relationships between several measurements of meat quality, including chemical and/or physical measurements and

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sensory evaluations, but the ability of simple physico-chemical measurements to differentiate rabbit meat has never been evaluated. The aim of the study was to establish whether the origins of breeding rabbits could be differentiated by simple physico-chemical measurements of the carcasses and meat. A second goal was to provide an objective and operating discriminating tool based on a few selected physico-chemical items. All types of measurements used in this study could be performed within a 24 h period because the final proposed system should be a useful tool to help trace the breeding origin of meat before commercialisation. For this purpose, we used three different breeding systems to produce high variability in meat quality: a standard production system, a breeding system complying with 'French' label norms, and a very slow growth rate breeding system. For each rearing system an appropriate genotype was used.

## MATERIALS AND METHODS

### Animals

At the experimental farm of ITAVI Rambouillet (France), three different rabbit breeding systems were settled according to animal strains, housing and feeding. The first two groups of rabbits were reared from weaning to slaughter according either to the standard intensive breeding system (group STAND;  $n = 102$ ) or to the 'French' label norms (group LABEL,  $n = 78$ ). The third group consisted of a particular rabbit breed (pure Himalayan, adult weight of 2.7 kg) characterised by very low growth rate (group RUSSE;  $n = 56$ ). Rabbits from the STAND group (PS Hyplus 19 × PS Hyplus 39, commercial hybrids, Grimaud Frères, France; adult weight 4.5 kg) were reared in collective cages of six animals at a stock density of 17.5 rabbits per m<sup>2</sup> and received a commercial pelleted feed *ad libitum*. Rabbits from the LABEL group (PS Hyplus 19 × PS Hyplus 99, commercial hybrids for the production of Label rabbits, Grimaud Frères France; adult weight 3.7 kg) were reared in pens of 36 animals at the same stocking density as rabbits from the STAND groups. Rabbits from the RUSSE group were reared in hutches of two to five animals (2.3 to 9.3 rabbits per m<sup>2</sup>). The LABEL and RUSSE rabbits were given, *ad libitum*, a commercial pelleted feed designed for Label production. Observed daily weight gains were  $42 \pm 6 \text{ g day}^{-1}$ ,  $28 \pm 8 \text{ g day}^{-1}$ , and  $15 \pm 11 \text{ g day}^{-1}$  for STAND, LABEL and RUSSE groups, respectively. Rabbits from the three groups were slaughtered at the same weight ( $2315 \pm 144 \text{ g}$ ) reached at 71 days, 92 days and 135 days for STAND, LABEL and RUSSE groups, respectively.

### Carcass and meat quality measurements

Animals were slaughtered without prior fasting and transportation and in compliance with French national regulations. After 24 h of chilling, the weighed carcass

was divided according to the recommendations of the World Rabbit Scientific Association.<sup>9</sup> Proportions of perirenal fat, interscapular fat, and fore- (WforeC,%), back- and hind-parts (WlegC,%) (weight/chilled carcass weight, ×100) were calculated. Retail cuts from the fore-, back- and hind-parts were vacuum-packed and frozen at  $-20^\circ\text{C}$ , until further analysis. Meat-to-bone ratio was determined in the leg (MBR).<sup>9</sup> The femur weight was expressed as percentage of hind leg weight (WLeg\_F,%) or of chilled carcass weight (WC\_F,%).

### Meat physico-chemical measurements

The day after slaughtering, ultimate pH was measured in muscle longissimus lumborum (LL, adjacent to the sixth lumbar vertebra level) and in biceps femoris (BF), using a combined glass penetrating electrode (Ingold, Mettler Toledo, Greifensee, Switzerland). Colour was assessed on the surface carcass over LL and BF and on a freshly exposed cut surface of LL. A Minolta CR-300 chromameter (Minolta, Osaka, Japan) was set to the  $L^*$  (lightness),  $a^*$  (redness), and  $b^*$  (yellowness) CIE scale ( $^{\circ}\text{L.LL}$  and  $^{\circ}\text{L.BF}$ ). After thawing, sample of LL was weighed, vacuum packed, cooked in a water bath ( $85^\circ\text{C}$ ) for 40 min and cooking loss was determined.<sup>10</sup> Water-holding capacity was estimated by centrifuging raw or cooked LL portions for 10 min at  $1500 \times g$ , and determining the residual water by drying the sample at  $103^\circ\text{C}$  overnight.<sup>11</sup> Moisture content was determined in raw and cooked LL (Mcr.LL and Mcc.LL,%) and dry matter in comestible part of fore and hind leg (DM.Fore DM.Leg,%) by drying at  $103^\circ\text{C}$  overnight. We used TOBEC methodology (total body electrical conductivity) on mixed deboned leg meat (E.Leg), mixed fore part (E.Fore) or entire LL as previously described.<sup>12</sup> TOBEC is a non-invasive technique that has been shown to accurately predict lean body mass or weight of total water in some mammals.<sup>13</sup> Briefly, the entire LL sample or gently centrifuged mixed meat (10 min,  $3000 \times g$ ) were placed in the middle of the detection chamber, under a 10 MHz oscillating magnetic field (EM-SCAN SA-3044 5EM SCAN Inc. Springfield, IL, USA). The energy loss was detected as a phase change in the impedance of the coil and expressed as the  $E$ -value.

### Loin meat mechanical properties

An entire cross-section in the mid-portion of raw or cooked LL was photographed and muscle area was measured by image analysis. Warner-Bratzler (WB) shear test was performed as previously described<sup>14</sup> using a WB device drawn at  $100 \text{ mm min}^{-1}$  adapted to a universal testing machine (Synergie 200, MTS, Eden Prairie, MN, USA). The LL muscle samples were positioned so that the superficial epimysial side was the last shared. The force displacement curve of raw LL has two peaks, the first corresponding to both myofibrils and endomysial collagen shear forces, and the second corresponding to the epimysial collagen shear force.<sup>15</sup> Due to the high heat solubility of

collagen of rabbit meat,<sup>10</sup> this latter peak is absent in the cooked meat. The recorded parameters from the force displacement curve were shear force applied at the first peak (F1r\_LL, N) at second peak (F3r\_LL, N) and at maximum whatever the peak (FMr\_LL, N). The level of the minimum force applied between the two peaks was recorded (F2r\_LL, N). Distances to those three points were also recorded. Energy was calculated as the area under the force displacement curve (TE\_LL, mJ). Stress was calculated according to Salé.<sup>16</sup>

#### *Bone shape and mechanical properties measurements*

Femurs were submitted to a three-point flexure test conducted with a universal testing machine (Synergie 200, MTS, Eden Prairie, MN, USA). Distance between the two fulcrum points supporting the bones was 30 mm and load was applied at 5 mm min<sup>-1</sup>. Length (length\_F, mm), and outside (b\_F and d\_F, mm) diameters at the point of loading, both perpendicular and parallel to the direction of the applied force, were measured using a dial calliper ( $\pm 0.02$  mm). The area moment of inertia (ML\_F, mm<sup>4</sup>), which is an estimation of bone distribution assuming that shape is similar to an elliptical plain tube was calculated according to the formula:  $MI = \pi \times (b_F \cdot d_F^3) / 64$ . Yield force (Y\_F, N), distance to yield force (DYF\_F, mm), energy to yield force (EYF\_F, mJ), ultimate force (UF\_F, N) and stiffness (slope of the elastic part : Stif\_F, N mm<sup>-1</sup>) were collected from the load deformation curve. Bone strain (Strain\_F) corresponding to the relative deformation of bone, maximum stress (StresUF\_F, N mm<sup>-2</sup>) defined as ultimate force per unit of bone area, and modulus of elasticity (Mod\_F, N mm<sup>-2</sup>) as a measure of the degree of bone rigidity,<sup>17</sup> were calculated according to formula reported by Patterson *et al.*<sup>18</sup>

#### **Data analysis**

Normal distribution of the residues was checked, and it was decided to transform six variables using the natural logarithm function. When applying multivariate analyses, observations with missing values are excluded. In the present study, only 192 rabbits out of 236 slaughtered had all measurements recorded. In order to keep all the rabbits in the analysis, when possible, missing values were replaced by a multiple imputed value using a Monte Carlo Markov chain method (MI procedure of SAS<sup>19</sup>). This procedure allows uncertainty on missing values to be taken into account, to fit the initial distribution and to keep the main relationships between variables. Quantitative values for the 63 variables measured in each of the 236 rabbits were first analysed by a one-way analysis of variance, including the group effect (PROC GLM of SAS<sup>19</sup>). From this analysis the most 30 relevant variables were kept, selected on their high  $R^2$  ( $R^2 > 0.18$  with  $P < 0.001$  for the group effect). Data were analysed in the following steps.

#### *Principal component analysis*

A principal component analysis (PCA) was performed using the 30 variables data set obtained after simple variance analysis ( $n = 224$ ) to provide a partial visualisation of the correlations between variables data set in a reduced dimension plot and also to allow a primary evaluation of the between-category similarity. Principal components (PCs) were calculated using the PRINCOMP procedure of SAS.<sup>19</sup>

#### *Linear discriminant analysis*

A stepwise linear discriminant analysis (LDA) was applied to the same 30 variables complete data set in order to obtain a reduced set of variables that best revealed the differences among the three groups, using the STEPDISC procedure of SAS.<sup>19</sup>

#### *Random division*

The data set was randomly divided between a learning (or training) set and an evaluation (or test) set, with 80% and 20% of initial observations, respectively. The random process applied for selecting the data was not constrained to respect the initial repartition of observations in each groups. The two linear combinations (also called first LDA1 and second LDA2 factors) that provide maximal separation between the groups were estimated (proc CANDISC of SAS<sup>19</sup>) on the learning set using the subset variables previously selected by STEPDISC.

#### *Decision tree analysis*

A decision tree analysis (DTA)<sup>20</sup> was applied on the learning set. The analysis was performed using the R 2.2.1 package.<sup>21</sup> DTA splits data into binary branches according to the values of variables and continues splitting branches in an iterative process that leads to the target value. Each split depends on the value of only one variable. Often the different suggested splits leads to extremely refined trees and thus to very unstable predictive models.<sup>22</sup> A procedure of tree pruning by cross-validation is then performed to keep good predictive performances and to allow a generalisation.

#### *Accuracy and the robustness*

The last step was performed to check the accuracy and the robustness of the variables selected by LDA or DTA procedures. The reliability of the LDA and DTA classification were checked both on learning and on evaluation sets. This final analysis was performed using the SAS DISCRIM procedure for the LDA and the R tree package for the DTA.

## **RESULTS AND DISCUSSION**

In our experiment, the main differences between the three groups are genetic background, housing system and feeding intensity. The combination of these factors allowed to produce rabbits with a physiological maturity (defined as the ratio between slaughter weight to

**Table 1.** Lsmeans of the 30 variables measured in carcass, meat and femur of rabbit reared in three different breeding systems. Variable were selected on their high  $R^2 > 0.18$  with  $P < 0.001$  for the group effect

Variable	STAND	LABEL	RUSSE	RMSE	$R^2$
<b>Conformation traits</b>					
WForeC (%)	30.16 <sup>b</sup>	31.68 <sup>a</sup>	32.16 <sup>a</sup>	1.52	0.250
WLegC (%)	14.79 <sup>b</sup>	15.55 <sup>a</sup>	14.44 <sup>c</sup>	0.91	0.189
MBR	6.01 <sup>b</sup>	6.16 <sup>b</sup>	7.51 <sup>a</sup>	0.60	0.509
<b>Colour traits of carcasses</b>					
*L.BF	55.18 <sup>a</sup>	51.89 <sup>b</sup>	52.44 <sup>b</sup>	2.67	0.249
*L.LL	56.17 <sup>a</sup>	53.15 <sup>b</sup>	52.94 <sup>b</sup>	2.53	0.273
<b>Water or lipid related traits</b>					
E.Fore	313 <sup>b</sup>	317 <sup>a</sup>	297 <sup>c</sup>	13	0.274
E.Leg	334.72 <sup>a</sup>	327.86 <sup>b</sup>	326.55 <sup>b</sup>	6.28	0.260
DM.Fore (%)	34.02 <sup>b</sup>	33.34 <sup>b</sup>	38.04 <sup>a</sup>	2.44	0.370
DM.Leg (%)	25.7 <sup>b</sup>	25.66 <sup>b</sup>	26.87 <sup>a</sup>	0.82	0.275
<b>Longissimus lumborum (LL) traits</b>					
M.Cr.LL (%)	74.29 <sup>a</sup>	73.72 <sup>b</sup>	72.75 <sup>c</sup>	0.72	0.417
M.Cc.LL (%)	66.69 <sup>a</sup>	65.07 <sup>b</sup>	64.53 <sup>c</sup>	0.93	0.203
Log(F1r.LL) (N)	3.50 <sup>b</sup>	3.73 <sup>a</sup>	3.75 <sup>a</sup>	0.17	0.322
F2r.LL (N)	25.16 <sup>b</sup>	33.32 <sup>a</sup>	34.90 <sup>a</sup>	6.01	0.353
Log(F3r.LL) (N)	3.44 <sup>c</sup>	3.76 <sup>b</sup>	3.90 <sup>a</sup>	0.21	0.450
FMr.LL (N)	35.10 <sup>c</sup>	45.82 <sup>b</sup>	51.36 <sup>a</sup>	8.61	0.385
TE.LL (mJ)	681 <sup>c</sup>	891 <sup>b</sup>	1021 <sup>a</sup>	174	0.396
<b>Femur traits and three-point flexure test</b>					
WC.F (%)	0.886 <sup>a</sup>	0.882 <sup>a</sup>	0.655 <sup>b</sup>	0.077	0.617
WLeg.F (%)	6.02 <sup>a</sup>	5.69 <sup>b</sup>	4.55 <sup>c</sup>	0.65	0.445
Length.F (mm)	80.57 <sup>b</sup>	85.31 <sup>a</sup>	80.15 <sup>b</sup>	2.02	0.568
b.F (mm)	8.14 <sup>a</sup>	8.03 <sup>a</sup>	7.60 <sup>b</sup>	0.43	0.204
d.F (mm)	6.54 <sup>b</sup>	6.73 <sup>a</sup>	6.04 <sup>c</sup>	0.31	0.408
Mi.F (mm <sup>4</sup> )	113 <sup>b</sup>	121 <sup>a</sup>	83 <sup>c</sup>	20	0.344
YF.F (N)	209 <sup>b</sup>	256 <sup>a</sup>	189 <sup>c</sup>	39	0.306
DYF.F (mm)	0.74 <sup>a</sup>	0.56 <sup>b</sup>	0.48 <sup>c</sup>	0.17	0.288
Log(EYF.F) (mJ)	4.36 <sup>a</sup>	4.27 <sup>a</sup>	3.77 <sup>b</sup>	0.36	0.305
UF.F (N)	290 <sup>c</sup>	398 <sup>a</sup>	311 <sup>b</sup>	44	0.549
StresUF.F (N mm <sup>-2</sup> )	64 <sup>b</sup>	84 <sup>a</sup>	86 <sup>a</sup>	12	0.434
Stif.F (N mm <sup>-1</sup> )	308 <sup>c</sup>	490 <sup>b</sup>	461 <sup>a</sup>	52	0.730
Mod.F (N mm <sup>-2</sup> )	1590 <sup>c</sup>	2331 <sup>b</sup>	3209 <sup>a</sup>	479	0.642
Strain.F	0.0322 <sup>a</sup>	0.0253 <sup>b</sup>	0.0193 <sup>c</sup>	0.0078	0.302

Abbreviations : WLegC = ratio between leg weight and chilled carcass weight time 100; WForeC = ratio between fore weight and chilled carcass weight time 100; MBR = meat-to-bone ratio; \*L.BF and \*L.LL = measure of lightness in biceps femoris and in longissimus lumborum (LL); E.Fore and E.Leg = TOBEC value measured in mixed fore part and mixed deboned leg; DM.Fore and DM.Leg = dry matter of mixed fore part and mixed deboned leg; M.Cr.LL and M.Cc.LL = moisture content of raw and cooked LL; F1r.LL, F2r.LL, F3r.LL and FMr.LL = Warner-Bratzler (WB) LL shear force value applied at the first peak, between 1<sup>st</sup> and 2<sup>nd</sup> peak, at the second peak and at maximum; TE.LL = WB total energy necessary to shear LL; WC.F = ratio between femur weight and chilled carcass weight time 100; WLeg.F = ratio between femur weight and leg weight time 100; Length.F = femur length; b.F and d.F = outside latero-medial and antero-posterior femur diameter; Mi.F = femur moment of inertia; YF.F = femur yield force; DYF.F = femur displacement at yield force; EYF.F = femur energy at the yield force; UF.F = femur ultimate force; StresUF.F = femur ultimate stress; Stif.F = femur stiffness; Mod.F = femur elastic modulus; Strain.F = femur strain. RMSE: root mean square error, (n = 236).

<sup>a,b,c</sup> Within a row, least squares means without a common superscript letter differ,  $P < 0.05$ .

adult weight) of 51, 63 and 85% for STAND, LABEL and RUSSE rabbits respectively. Table 1 shows the 30 variables selected ( $R^2 > 0.18$ ,  $P > 0.001$  for group effect) among the 63 variables initially measured or assessed by computation, and associated  $R^2$ . Interestingly, pH, water holding capacity, cooking loss values and variables collected after Warner-Bratzler shear test on cooked longissimus lumborum (LL) muscle were not able to discriminate compared groups in our experiment. In agreement, previous experiments have shown that shear test parameters allow a clear differentiation between rabbit meats of LABEL production or standard breeding system when performed on raw

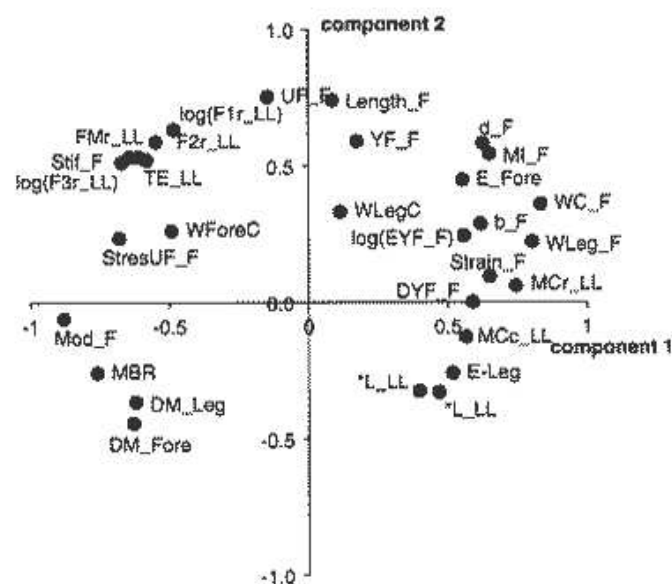
but not on cooked LL.<sup>12</sup> Based on Lsmeans comparison, only 16 out of 30 variables allowed a clear discrimination between the three groups. Nine corresponded to bone traits (shape measurement and three points-flexure test), and five to LL characteristics (i.e. water content and Warner-Bratzler test on raw sample). The rank order observed for the three groups for water content of raw or cooked LL,<sup>23-25</sup> toughness of raw LL<sup>25</sup> and bone elasticity<sup>25</sup> may partly be attributed to differences in rabbit age at the same body weight at slaughter. The highest leg proportion (WLegC) observed in LABEL rabbit compared to STAND rabbits may be related to spontaneous physical exercise

allowed by pen housing compared to caged housing, since it has been regularly observed in other studies on space allowance for rabbits.<sup>26–29</sup>

In order to have an overview of relationships between these 30 variables as well as of differences between groups, a PCA analysis was performed. The first four PC explained 69% of total variation (Table 2). Figure 1 shows a plot of the different traits according to the first two PC. A first group of variables, which was highly negatively correlated to the first PC (Table 3), included meat-to-bone ratio (MBR) as

**Table 2.** Eigen values, explained variance and cumulative variance of the four first principal components (PCs) of principal component analysis

	Eigen value	Explained variance	Cumulative variance
PC1	10.45	34.86	34.86
PC2	5.40	18.00	52.86
PC3	2.62	8.76	61.62
PC4	2.33	7.29	69.39

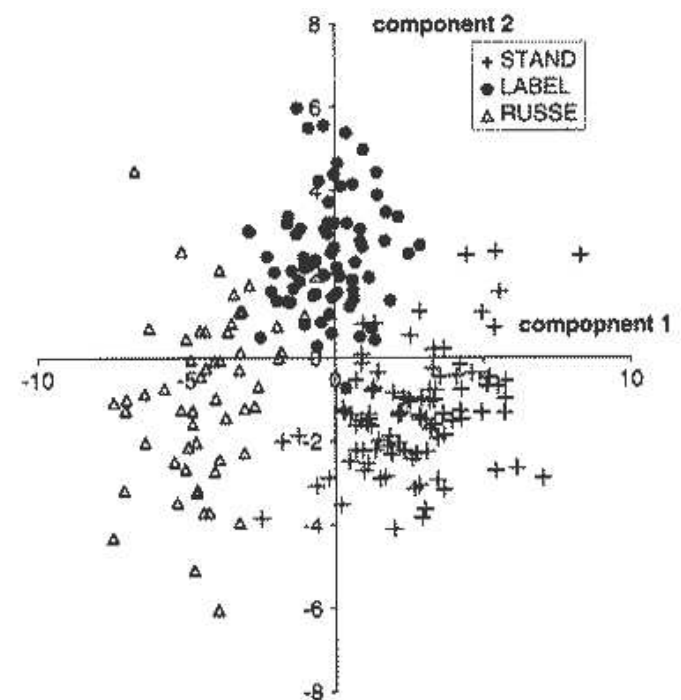


**Figure 1.** Projection of the 30 variables (selected on their high  $R^2 > 0.18$  with  $P < 0.001$ ) on the plane defined by the first two principal components (component1 and component2). Abbreviations: WLegC = ratio between leg weight and chilled carcass weight  $\times 100$ ; WforeC = ratio between fore weight and chilled carcass weight  $\times 100$ ; MBR = leg meat to bone ratio; \*L<sub>BF</sub> and \*L<sub>LL</sub> = measure of lightness in biceps femoris and in longissimus lumborum (LL); E<sub>Fore</sub> and E<sub>Leg</sub> = TOBEC value measured in mixed fore part and mixed deboned leg; DM<sub>Fore</sub> and DM<sub>Leg</sub> = dry matter of mixed fore part and mixed deboned leg; MCr<sub>LL</sub> and MCo<sub>LL</sub> = moisture content of raw and cooked LL; F1r<sub>LL</sub>, F2r<sub>LL</sub>, F3r<sub>LL</sub> and FMr<sub>LL</sub> = Warner-Bratzler (WB) LL shear force value applied at the first peak, between the first and second peaks, at the second peak and at maximum; TE<sub>LL</sub> = WB total energy necessary to shear LL; WC<sub>F</sub> = ratio between femur weight and chilled carcass weight  $\times 100$ ; Wleg<sub>F</sub> = ratio between femur weight and leg weight  $\times 100$ ; Length<sub>F</sub> = femur length; b<sub>F</sub> and d<sub>F</sub> = outside latero-medial and antero-posterior femur diameter; MI<sub>F</sub> = femur moment of inertia; YF<sub>F</sub> = femur yield force; DYF<sub>F</sub> = femur displacement at yield force; EYF<sub>F</sub> = femur energy at the yield force; UF<sub>F</sub> = femur ultimate force; StresUF<sub>F</sub> = femur ultimate stress; Stif<sub>F</sub> = femur stiffness; Mod<sub>F</sub>: femur elastic modulus; Strain<sub>F</sub>: femur strain.

**Table 3.** Loading of variables in the first two principal components (PC1 and PC2)

Variable	PC1	PC2	Variable	PC1	PC2
Mod <sub>F</sub>	-0.880	-0.062	YF <sub>F</sub>	0.175	0.590
MBR	-0.759	-0.260	*L <sub>BF</sub>	0.398	-0.326
StresUF <sub>F</sub>	-0.680	0.235	*L <sub>LL</sub>	0.468	-0.330
Log(F3r <sub>LL</sub> )	-0.670	0.511	E <sub>Leg</sub>	0.516	-0.258
Stif <sub>F</sub>	-0.642	0.532	E <sub>Fore</sub>	0.552	0.449
DM <sub>Fore</sub>	-0.630	-0.444	Log(EYF <sub>F</sub> )	0.558	0.243
DM <sub>Leg</sub>	-0.621	-0.366	MCo <sub>LL</sub>	0.565	-0.127
FMr <sub>LL</sub>	-0.609	0.532	DYF <sub>F</sub>	0.589	0.001
TE <sub>LL</sub>	-0.580	0.521	b <sub>F</sub>	0.618	0.288
F2r <sub>LL</sub>	-0.547	0.588	d <sub>F</sub>	0.627	0.583
WForeC	-0.492	0.261	MI <sub>F</sub>	0.650	0.544
Log(F1r <sub>LL</sub> )	-0.482	0.632	Strain <sub>F</sub>	0.650	0.094
UF <sub>F</sub>	-0.145	0.753	MCr <sub>LL</sub>	0.745	0.060
Length <sub>F</sub>	0.086	0.740	WLeg <sub>F</sub>	0.802	0.220
WLegC	0.114	0.333	WCarc <sub>F</sub>	0.834	0.359

Abbreviations are as given in Table 1.



**Figure 2.** Projection of the data of the three groups of rabbit (STAND +, LABEL ●, and RUSSE Δ) on the plane defined by the first two principal components.

a conformation variable, bone modulus of elasticity (Mod<sub>F</sub>) as an indication of basic material elasticity independent of geometry, bone stress (StresUF<sub>F</sub>) as a measure of force per unit of bone area, femur stiffness (Stif<sub>F</sub>), and mechanical toughness of raw LL (log(F3r<sub>LL</sub>), FMr<sub>LL</sub> and TE<sub>LL</sub>). This group was opposed to a second group of conformation variables (WLeg<sub>F</sub> and WC<sub>F</sub>), to moisture content of raw and cooked LL (MCr<sub>LL</sub>, MCo<sub>LL</sub>), to femur strain (Strain<sub>F</sub>) and to three variables related to bone shape (MI<sub>F</sub>, b<sub>F</sub> and d<sub>F</sub>). The second PC was essentially related to bone characteristics (UF<sub>F</sub>, Length<sub>F</sub>, YF<sub>F</sub>). Figure 2 shows the projection of data on the first two PC. The first PC opposed STAND and

RUSSE rabbits, suggesting that RUSSE rabbits were characterised by higher bone rigidity, lower moisture content and higher toughness of raw LL than STAND rabbits. LABEL rabbits had an intermediate position for the later parameters. The second PC was related to LABEL rabbits opposed to non-discriminated STAND and RUSSE animals, suggesting that LABEL animals had the longest femur with the highest yield and ultimate flexure forces. PCA has previously been used to describe meat quality in rabbit<sup>7,8</sup> but with a limited number of physico-chemical (or sensorial) measurements. To our knowledge, bone and muscle mechanical characteristics have never been evaluated in PCA. Considering the present results, it seems that bone mechanical characteristics might play an important part in describing the variation observed in carcasses of rabbits produced according to different breeding systems allowing different growth rate.

The stepwise linear discriminant analysis selected 14 variables among the 30 variables, as the best discriminant traits between the three groups. Their respective coefficients for the two LDA factors are reported in Table 4. The explained percentage of variance for the first factor was 60.9%. The graphic representation of the LDA (Fig. 3) clearly illustrates the ability of the 14 variables to discriminate the three groups. Only eight variables out of 14 corresponded to LSmeans selected variables. Eight variables were related to bone traits, and five out of eight corresponded to mechanical femur parameters. The first two variables were related to bone mechanical

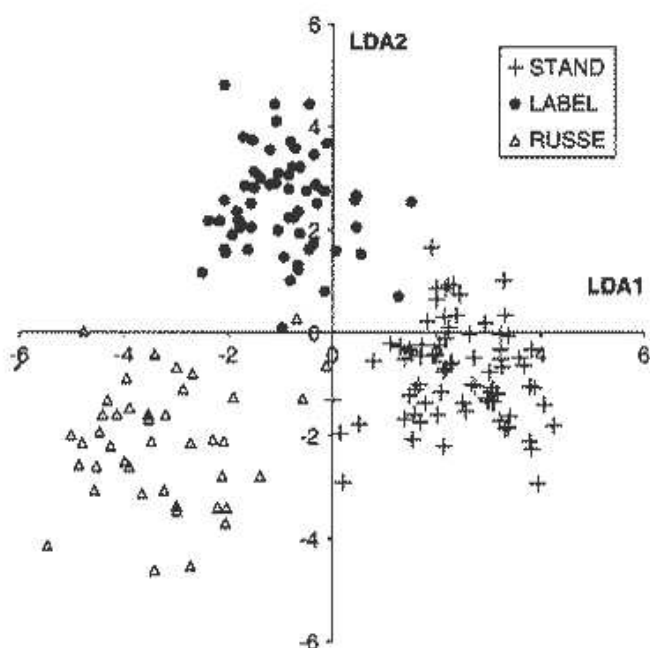
**Table 4.** First and second factor (LDA1 and LDA2) coefficients and the explained percentage of variance of linear discriminant analysis in decreasing order of their contribution to the first LDA factor

Rank order	Variable	LDA1	LDA2
1	Stif.F	-0.0030	-0.0024
2	UF.F	-0.0028	0.0078
3	TE.LL	-0.0012	-0.0016
4	Length.F	-0.0820	0.1500
5	Mod.F	0.3800	0.5570
6	MI.F	-0.0524	-0.0394
7	*L.BF	0.0743	-0.1109
8	log(EYF.F)	1.5185	1.4390
9	DYF.F	-2.0480	-3.5801
10	log(F1r.LL)	-1.1450	1.0070
11	WLegC	-0.1113	0.1118
12	WLeg.F	0.8566	0.9633
13	WForeC	0.0041	0.0149
14	E.Leg	0.0148	-0.0211
Explained % of variance		60.91	39.09

Abbreviations are given in table 1

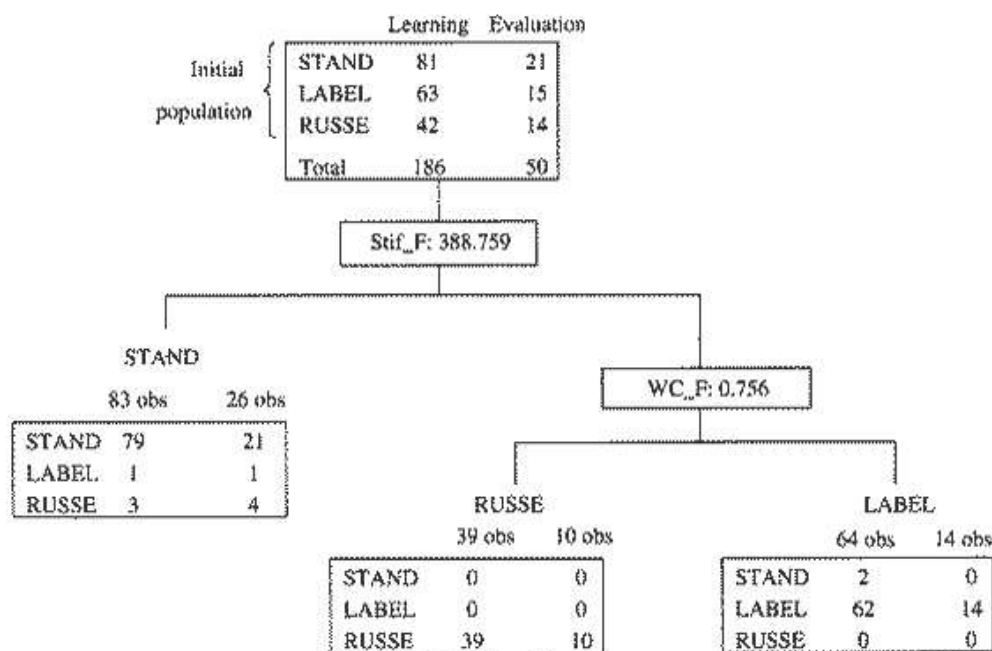
**Table 5.** Percentage of correctly classified rabbits and misclassified rabbits from the three groups using LDA method

	Correct assignment (%)	STAND rabbit misclassified (%)	LABEL rabbit misclassified (%)	RUSSE rabbit misclassified (%)
Learning set	98.35	0.00	1.64	4.76
Evaluation set	96.00	0.00	6.67	7.14



**Figure 3.** Canonical representation of the three groups of rabbits (STAND +, LABEL •, and RUSSE Δ) on the plane given by the two linear discriminant factors.

characteristics, i.e. the slope of the elastic region from load deformation curve (Stif.F) and ultimate force applied to femur (UF.F). Mechanical characteristics of LL muscle appeared at the third and tenth position. The recognition ability of LDA was satisfactory since 98.3% and 96.0% of rabbits were correctly assigned to their original group for the learning and the evaluation sets, respectively (Table 5). The LDA, especially, allowed an accurate classification of STAND rabbits, because none was misclassified. The misclassification rate of LABEL group was low, with 1.6% and 6.7% for the learning and the evaluation sets, respectively. Indeed, only one LABEL rabbit out of the 61 analysed was misclassified in the STAND group. In the evaluation set, one LABEL rabbit out of 15 was misclassified in STAND group. The worst classification rate was encountered for RUSSE rabbits (4.8 and 7.1% for the learning and evaluation sets, respectively). In the learning set, two RUSSE rabbits out of 42 were assigned to the STAND and to the LABEL groups, respectively, while one RUSSE rabbit out of 14 was assigned to LABEL group in the evaluation set. Consequently, these 14 physico-chemical traits can be very efficiently used for discriminating rabbits from different breeding origins. However, measurements to be recorded for accurate classification of rabbit meat might be still considered as too numerous in a commercial context. Indeed to



**Figure 4.** Decision tree analysis performed on learning set (left column) and evaluation set (right column). Femur stiffness (Stif.F) was selected as the first splitting variable to discriminate STAND group from the two other groups, whereas use of ratio between femur weight and leg weight  $\times 100$  (WLeg.F) was selected as second splitting variable and allow to separate RUSSE from LABEL in the group of rabbits with a femur rigidity higher than  $393.93 \text{ N mm}^{-1}$ . In the learning set, two STAND rabbits out of 81 were misclassified in the LABEL group. For both the learning and evaluation sets, one LABEL was classified in the STAND group. In the learning set, three RUSSE rabbits out of 42 were assigned to the STAND group, while in the evaluation set, four RUSSE rabbits out of 14 were assigned to the STAND group.

**Table 6.** Percentage of correctly classified rabbits and misclassified rabbits from the three groups using the DTA method

	Correct assignment (%)	STAND rabbit misclassified (%)	LABEL rabbit misclassified (%)	RUSSE rabbit misclassified (%)
Learning set	96.77	2.47	1.59	7.14
Evaluation set	90.00	0	6.67	28.57

assess those 14 variables, it is necessary to perform six different measurement methods and to have the whole carcass available.

In decision tree analysis, there is a balance between accuracy (no classification errors) and robustness (lower number of leaves obtained by pruning). The final decision tree was the most simple with three remaining leaves (Fig. 4). Then, DTA selected the same first variable as LDA, i.e. the slope value of the elastic part of the three points-flexure test (Stif.F). A cut-off value of  $388.8 \text{ N mm}^{-1}$  was set up for bone stiffness, with rabbits with femur rigidity under this value classified as STAND. For  $\text{Stif.F} > 388.8 \text{ N mm}^{-1}$ , the ratio of femur weight to chilled carcass weight (WC.F) was defined as the second splitting variable. This latter variable was not selected by LDA, but was highly correlated ( $R^2 = 0.88$ ) to the ratio of femur weight to hind leg weight (WLeg.F, i.e. the 12th variable selected in LDA). Among rabbits whose femur rigidity was over  $388.8 \text{ N mm}^{-1}$ ,  $\text{WC.F} < 0.756\%$  classified RUSSE group while  $\text{WC.F} > 0.756\%$  classified LABEL group. Using DTA, 96.8% and 90.0% of rabbits were correctly assigned to their group for the learning and evaluation sets respectively (Table 6). The misclassification rate was 2.5% and 0% for STAND rabbits for learning and

evaluation sets, respectively. The misclassification rate in the LABEL group was 1.6% and 6.7% for learning and evaluation sets, respectively. The highest misclassification rate was encountered for RUSSE rabbits (7.1% and 28.6% for learning and evaluation sets, respectively). Unlike LDA, DTA variables can be assessed using two different measurement methods and required only a leg retail cut.

In the present study, LDA and DTA systems were developed to discriminate rabbit meat. Thus the physico-chemical measurements chosen were specific to this species. Nevertheless, these systems could be used for animals bred in large batches such as poultry. Indeed, in both discrimination systems, the most relevant variables were related to bone mechanical characteristics. It has been shown previously<sup>30</sup> that bone mechanical characteristics might play an important part in describing the variation observed in carcasses of chicken produced according to different rearing systems allowing different growth rates.

## CONCLUSION

This work demonstrated that simple physico-chemical traits recorded in carcasses and meat were efficient

to discriminate rabbits from three different productive processes. Mechanical characteristics of femur and loin muscle explained much of the total variability. Two different tools of discrimination were constructed using LDA and DTA procedures. Although the recognition ability of the three groups was higher for LDA than for DTA, DTA discrimination system is far simpler than LDA and enlightened only two variables. These systems could have further implications for future traceability of breeding origin. The DTA provides a simple system of discrimination to be prospectively tested.

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