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#### MICROBIAL DETERIORATION OF LAMB MEAT OF PORTUGUESE ORIGIN AS AFFECTED BY ITS INTRINSIC PROPERTIES

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### Introduction

- In Portugal, there are 15 autochthonous sheep breeds
- The main autochthonous sheep breeds exploited for meat production are Churra-Galega-Bragançana (CGB) and Bordaleirade-Entre-Douro-e-Minho (BEDM)
- Sheep and goat meat production constitutes 2.8% of Portugal's meat production



Galega Bragançana

Figure 1. Churra Galega Bragançana Mediterrânea (Bragança)



Figure 2. Bordaleira-entre-Douro-E-Minho Atlântica (Ponte de Lima)





# Introduction

- Lamb meat achieves ~80% of its maximum tenderness potential after 7 days of cold maturation
- During maturation, microbial deterioration takes place due to the proliferation of psychrotrophic bacteria, lactic acid bacteria, *Pseudomonas* spp., *Clostridium* spp., etc.
- Vacuum packaging (VP) is well-known to prolong the life of the meat, by retarding microbial deterioration. It depends on:
  - chilling system/profile
  - initial microbial contamination
  - physicochemical or intrinsic properties of the meat



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Figure 3. Lamb carcasses







i) To evaluate the evolution of spoilage indicator microorganisms in refrigerated vacuum-packed (VP) lamb meat from two Portuguese breeds, Churra-Galega-Bragançana (CGB) and Bordaleira-de-Entre-Douro-e-Minho (BEDM)

ii) To elucidate any interrelationship between meat's intrinsic properties (i.e., pH, water activity and proximate composition) and microbial growth



### **Sample population**

- 30 CBG and 30 BDEM lambs
- Lambs reared in autumn 2018 and spring 2019
- Slaughtered at 4 months of age



Galega Bragançana





Entre-Doura e.



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- After carcass splitting, the left side of the Longissimus dorsi muscle was removed from the 6th to the 13th vertebra under aseptic conditions, and was vacuum-packed for microbiological analysis
- The right half of the muscle was kept for physicochemical analyses



Figure 5. Longissimus dorsi muscle



**Figure 6.** Scheme of the division of the *L. thoracis* and *L. lumborum* muscles for analysis







### **Physicochemical analysis:**

On Day 1

- √рН
- ✓ Water activity  $(a_w)$
- ✓Moisture content
- ✓ Fat content
- ✓ Protein content
- $\checkmark$  Ash content

### **Microbiological analysis:**

### On Days 3, 9 and 15

- ✓ Total viable counts
- ✓ Psychrotrophic bacteria
- ✓ Lactic acid bacteria
- ✓ Pseudomonas spp.





#### **Statistical analysis**

• Mixed effects models: Aimed to understand to what extent the intrinsic or physicochemical properties of meat (i.e., pH, aw and proximate composition) can affect the growth of mesophiles, lactic acid bacteria, psychrotrophic bacteria and *Pseudomonas* spp.





- The **controlled hygiene process of the abattoir ensured relatively low bacterial counts** in meat, even on day 3 post-slaughter
- The lowest initial microbial populations were found for Pseudomonas and LAB
- Microbial populations of CGB meat were significantly lower for all microbial groups than in BEDM meat



Figure 7. Increase in mesophiles and lactic acid bacteria

Figure 8. Increase in *Pseudomonas* spp. and psychrotrophic bacteria



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- Significant effect of 'Breed' on mesophiles, LAB, Pseudomonas and psychrotrophic bacteria counts
- The effect of pH on microbial growth was significant for all bacterial groups, as observed in the interaction term pH×Day (mesophiles, p=0.006; LAB, p=0.001; Pseudomonas, p=0.050; psychrotrophic bacteria, p=0.010)

Model	Variable	Mesophiles	LAB	Pseudomonas	Psychrotrophic
рН	Breed	7.503 (0.008)	17.70 (<.0001)	8.147 (0.006)	6.100 (0.017)
	Day	168.4 (<.0001)	56.99 (<.0001)	32.15 (<.0001)	159.1 (<.0001)
	pH×Day	7.951 <b>(0.006)</b>	12.24 <b>(0.001)</b>	3.795 <b>(0.050)</b>	6.893 <b>(0.010)</b>
	Breed	7.259 (0.009)	18.32 (<.0001)	8.520 (0.005)	6.308 (0.015)
a <sub>w</sub>	Day	159.9 (<.0001)	50.79 (<.0001)	31.28 (<.0001)	148.7 (<.0001)
_	a <sub>w</sub> ×Day	0.050 (0.823)	0.211 (0.646)	3.082 (0.082)	1.019 (0.315)
	Breed	10.16 (0.002)	20.02 (<.0001)	8.854 (0.004)	7.141 (0.009)
Moisture	Day	169.5 (<.0001)	53.86 (<.0001)	35.53 (<.0001)	178.5 (<.0001)
	Moisture×Day	28.97 <b>(&lt;.0001)</b>	12.23 <b>(&lt;.0001)</b>	21.00 <b>(&lt;.0001)</b>	30.76 <b>(&lt;.0001)</b>

**Table 1.** Effects of lamb breed and initial intrinsic factors of meat on the concentration of spoilage microorganisms in refrigerated vacuum-packed meat as quantified by mixed models (F-values and p-values from analysis of variance are shown.



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**Figure 9.** Box plots of lamb *Longissimus dorsi* pH measured 24 hourspost-slaughter, by breed, Churra-Galega-Bragançana (CGB) and Bordaleira-entre-Douro-e-Minho (BEDM)

- Lower microbial populations in CGB lamb meat is linked to lower ultimate pH (5.58 for CGB vs. 5.77 for BEDM)
- The rapid depletion of glycogen levels, prompted by stress, prevents the normal drop in pH to optimal levels. As a result, meat of higher pH (>5.7) has better conditions for microbial growth, ultimately leading to a reduction of shelf-life in refrigerated conditions even when vacuum-packaging is applied



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- Water activity of meat had no effect on microbial growth, as seen by the non-significance of a<sub>w</sub>×Day (p values from 0.082 to 0.823)
- The development of deteriorating bacteria was found to be exacerbated by the moisture content of meat (p<.0001 for all interactions Moisture×Day)</li>

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**Table 1.** Effects of lamb breed and initial intrinsic factors of meat on the concentration of spoilage microorganisms in refrigerated vacuum-packed meat as quantified by mixed models (F-values and p-values from analysis of variance are shown.







 Lamb meat samples of higher intramuscular fat content underwent a slower microbial deterioration, as implied by the negative interaction Fat×Day (not shown) that was significant for all microbial groups (p<.0001)</li>

Model	Variable	Mesophiles	LAB	Pseudomonas	Psychrotrophic
Fat	Breed	9.314 (0.004)	19.75 (<.0001)	8.754 (0.005)	6.973 (0.011)
	Day	184.67 (<.0001)	56.73 (<.0001)	36.00 (<.0001)	185.5 (<.0001)
	Fat×Day	33.30 <b>(&lt;.0001)</b>	17.84 <b>(&lt;.0001)</b>	22.01 <b>(&lt;.0001)</b>	34.51 <b>(&lt;.0001)</b>
Protein	Breed	7.243 (0.009)	18.32 (<.0001)	8.196 (0.006)	6.192 (0.016)
	Day	161.9 (<.0001)	52.31 (<.0001)	31.59 (<.0001)	152.2 (<.0001)
	Protein×Day	1.392 (0.241)	3.612 (0.060)	2.085 (0.151)	2.576 (0.111)
	Breed	9.047 (0.004)	18.74 (<.0001)	8.591 (0.005)	6.629 (0.013)
Ashes	Day	172.0 (<.0001)	56.67 (<.0001)	33.11 (<.0001)	166.1 (<.0001)
	Ash×Day	22.25 <b>(&lt;.0001)</b>	14.67 <b>(0.001)</b>	10.32 <b>(0.002)</b>	16.87 <b>(0.001)</b>

**Table 1 (cont).** Effects of lamb breed and initial intrinsic factors of meat on the concentration of spoilage microorganisms in refrigerated vacuum-packed meat as quantified by mixed models (Fvalues and p-values from analysis of variance are shown.



- Although protein content of lamb meat presented a wide range of variation (80.97-91.34% (db)), protein content was not found to regulate the growth of spoilage bacteria in VP lamb meat (Protein×Day was non-significant in all bacterial groups)
- On the contrary, lamb meat samples of higher content of metal salts and trace minerals (ashes) tended to have a faster microbial spoilage (p of at least 0.001)

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### Conclusions



✓ Populations of spoilage bacterial groups were higher in VP lamb meat from BEDM breed than in that of CGB breed

✓ Because pH was significantly higher in BEDM lamb meat.

✓ A high ultimate pH was demonstrated to increase the rate of microbial deterioration

✓ High moisture and ash content in meat also increased the rate of microbial spoilage





### Conclusions

- ✓A higher intramuscular fat content tended to delay microbial spoilage
- ✓ In order to extend the shelf-life of Portuguese-origin lamb meat, animal handling and carcass classification can be improved towards the selection of fatter animals and chilled carcasses of optimal ultimate pH



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