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# Effects of stress by unfamiliar sounds on carcass and meat traits in bulls from three continental beef cattle breeds at different ageing times



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#### ABSTRACT

One-hundred-and-twenty-four young bulls of three Continental beef cattle breeds were used to study the effect of pre-slaughter stress by unfamiliar sounds on carcass traits and quality characteristics of beef aged for three different periods. Stress due to unfamiliar noises had a moderately negative effect on carcass and meat quality. Carcasses from group Stressed had higher (P < 0.05) values of pH<sub>0</sub>, pH<sub>24</sub>,  $a^*$ , and Warner-Bratzler shear force, and lower (P < 0.05) values of  $L^*$ ,  $b^*$  and pressure losses than from Unstressed animals. There were significant differences (P < 0.05) among breeds in  $L^*$ ,  $a^*$  and pressure losses. The ageing time had a significant effect on chromatic variables, WBSF and pressure losses. The cooking losses were not significantly affected by any of the three factors discussed. These results emphasize the importance of implementing appropriate management practices during pre-slaughter handling of cattle in order to reduce any possible risk factor for stress, as well as the different sensitivity of the breeds to similar stimuli.

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# 1. Introduction

The tolerance for alterations in environmental homeostasis has received greater interest in recent years due to the relationship with meat quality (Voisinet, Grandin, O'Connor, Tatum, & Deesing, 1997). Alterations to animal's homeostasis can happen at different times and places, such as the slaughter facilities where unfamiliar noises could act as a stressor. Cattle destined for meat production spend either a few hours or a whole night at a slaughterhouse to recover from the physical and psychological stress caused by transport and to achieve an acceptable muscle glycogen concentration at slaughter, which is one of the most important factors affecting beef quality. However, holding the cattle in lairage involves exposure to acute stressors (Ferguson & Warner, 2008), such as noise. The effect of the stressors depends on their type and number (stressors appear to be additive, Bray, Graafhuis, & Chrystall, 1989), their duration and intensity, the susceptibility of the animal to them (Ferguson, Shaw, & Stark, 2007), and the breed (Muchenje, Dzama, Chimonyo, Strydom, & Raats, 2009).

The design of the slaughter facilities has a strong relationship with the number and intensity of stressful stimuli (Grandin, 1990), and thus with stress-related meat quality problems, such as high pH, abnormal color, and tough meat. In some European abattoirs, the holding area has pens for different species (pig, cattle, sheep, goat) from which they are not seen but heard. Pigs are very noisy during

pre-slaughter handling, while cattle have sensitive hearing and they are easily stressed (Waynert, Stookey, Schwartzkopf-Genswein, Watts, & Waltz, 1999). On the other hand, cattle with excitable temperaments could be more susceptible to stress caused by handling, and consequently have consistently poorer objective meat quality characteristics (Cafe et al., 2011; Voisinet et al., 1997).

The behavioural and physiological responses to noises have been studied in dairy cattle (Arnold, Ng, Jongman, & Hemsworth, 2007), while relatively few published studies have described relationships between noises in the pre-slaughter hours and commercially important traits of carcass and meat quality (Ferguson & Warner, 2008; Waynert et al., 1999). Therefore, the aim of the current work was to study the influence of pre-slaughter stress by unfamiliar noises during lairage period on carcass and meat quality parameters such as pH, water holding capacity, colour and shear force in Charolais, Limousine and Retinta bulls. Ageing times were set to mimic the ageing times used in the literature.

#### 2. Materials and methods

#### 2.1. Animals

The study animals included one-hundred twenty-four young bulls of three Continental beef cattle breeds (42 Charolais, CH; 42 Limousine, LI; and 40 Retinta, RE) born in different farms in the region of Andalucía (Spain), sired by one of fifteen different bulls. At seven-eight months of age they were weaned and moved to a commercial feedlot (average

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 $219\pm20.4$  days of age and  $252\pm25.4$  kg) located in south-western Spain (Córdoba). In the feedlot, the bulls were divided into six groups (two groups per breed) and assigned to feedlot pens (10  $\times$  10 m, 20–22 head per pen). Bulls were managed according to Spanish rules and regulations for animal care (EU Council Directive 86/609/EEC) and fattened under identical feeding and handling conditions. Feeding was ad libitum with a medium energy diet based on concentrate meal and wheat straw to promote good growth. When the animals reached the target weight (500–525 kg for RE, and 540–580 kg for CH and LI, according to local market preferences, average 418  $\pm$  28.8 days of age and  $534.9\pm27.5$  kg), they were transported to an officially approved abattoir (approximately 30 min and 20 km from feedlot) the afternoon before to slaughter. Travel conditions and handling were the same for all young bulls.

#### 2.2. Lairage groups and slaughtering procedures

The study was carried out in winter, when Iberian pigs and cattle can be held simultaneously in the lairage for subsequent slaughter. The bulls were allocated into two treatments, according to their environmental conditions during the lairage period prior to slaughter, with 3 replicates in three different days for each treatment (6–7 bulls of each breed per replicate, resulting in three groups of data for each breed and treatment). The treatments included Unstressed group (63 bulls: 22 CH, 21 LI, and 20 RE) and Stressed group (61 bulls: 20 CH, 21 LI, and 20 RE). On arrival at the slaughterhouse the afternoon before the date of slaughter, each sub-lot of animals of the same breed were kept in collective pens (concrete floor and metallic fencing) in front of the pig pens. There was one pen per each breed (with access to water but not food), and the animals were not mixed with others bulls from different backgrounds. The bulls were rested by isolating them from human activity, although auditory and visual contact could be established with other resting pens. Animals from Stressed group were slaughtered on days in which the lairage contained pigs (slaughtered at 4 to 7 am), while the animals of Unstressed group did not coincide with pigs in the lairage. The bulls were weighed and slaughtered at 8-10 am, approximately 12 h after arrival. All animals were handled, stunned by captive bolt and dressed according to European regulations for the Protection of Animals at Slaughter or Killing (93/119/EC D, 1993).

## 2.3. Carcass and meat quality analyses

Carcasses were weighed immediately after slaughter and a trained and experienced technician graded carcasses for conformation (with a 18-point scale: 1 = Poor, 18 = Superior +) and fatness (scale 1-5 point, being 5 the highest fatness) according to the European beef grading system (Regulation No 1208/81, OJEC, 1981, and its amending Regulation No 1026/91, OJEC, 1991). Then, the carcasses were stored in a chilling room. The pH, temperature and color of carcasses were recorded at the end of the slaughter line (approximately forty five min post-mortem), and after the chilling process (at 2-4 °C for 24 h). Carcass pH and temperature were measured on the Longissimus thoracis muscle at the level of the 13th thoracic vertebra of the right side, at right angles to the sagittal plane surface, with a portable pH-meter (Hanna HI9025) equipped with a glass electrode suitable for meat penetration and automatic temperature compensator. Carcass and meat color ( $L^* = \text{lightness}$ , a measure of the light reflected (100 = white; 0 = black);  $a^*$  = redness, measures positive red and negative green; and  $b^* =$  yellowness, measures positive yellow, negative blue) was performed using a CM-2600d spectrophotometer (Minolta Co., Osaka, Japan) (illuminant: D65; visual angle: 10°; measurement aperture: 8 mm). Colour was measured on the cutting surface of the m. Rectus abdominis and m. Longissimus thoracis. Three different locations were scanned and averaged for statistical analyses. Calibration was performed by using standard white tiles ( $L^* = 97.47$ ,  $a^* = -0.20$  and  $b^* = 1.79$ ) prior to the colour measurement. Colour coordinates were expressed as  $L^*$ ,  $a^*$  and  $b^*$ , following the  $L^*a^*b^*$  system (CIE, 1986). After chilling (24 h after slaughter), a section of about 21 cm was removed by a complete cross-section of the m. *Longissimus thoracis* from the left side of each carcass between ninth and thirteenth thoracic vertebrae. Samples were immediately vacuum packaged and transferred to the laboratory in refrigerator for the corresponding analysis. Thereafter, the loin muscle samples of each carcass were further divided into three subsamples (T10, T11, and T12: tenth, eleventh and twelfth thoracic vertebrae). T10 samples (aged for 1 day) were immediately analyzed. The rest of the subsamples were packaged in vacuum bags, aged for 7 (T11) and 21 (T12) days at 2–4 °C, and then analyzed (Fig. 1).

The meat quality characteristics (color, water holding capacity, Warner-Bratzler shear force) were recorded at 1, 7 and 21 days of ageing time. In order to assess meat color and water-holding capacity (WHC) from raw meat, a 2 cm length of Longissimus thoracis muscle was cut from the caudal end of each rib (Fig. 1). The fresh cut samples were displayed on trays, covered with plastic film permeable to oxygen and stored for 1 h at 4 °C for color measurement. Then, the color of fresh meat was measured using a CM-2600d spectrophotometer (Minolta Co., Osaka, Japan) (illuminant: D65; visual angle: 10°; measurement aperture: 8 mm). Three measurements were taken from each sample, and averaged for statistical analyses. The WHC was determined as pressure loss (PL) using a modified Grau and Hamm method described by Beriain et al. (2000), and expressed as press juice (the per cent of expelled juice after compression of 5 g meat samples with 2250 g applied for 5 min. A 5 cm length of Longissimus thoracis muscle was cut from the cranial end of each rib (Fig. 1) to assess Warner-Bratzler shear force (WBSF) and cooking loss (CL). Meat samples were cooked at 190 °C on a double plate grill until they reached an internal temperature of 70 °C. Internal temperature of the cooked muscle was monitored by an iron/constantan thermocouple wire connected to a thermometer and inserted into the geometric center of the sample. After cooking, the samples were cooled at room temperature for 30 min and the percentage of cooking loss recorded. CL was calculated as the percent weight difference between fresh and cooked samples (after gently blotting on filter paper) relative to the weight of fresh samples using the following equation: [(raw weight-cooked weight)/raw weight]\*100. Then, six 1.27 cm diameter cores per steak were removed parallel to muscle fibre orientation from the lateral end of the cooked steaks. Peak WBSF was measured perpendicular to the muscle fibres using a Texture Analyser (Model TA.XT-2, Texture Analyser® (Stable Micro Systems, Surrey, UK) equipped with a Warner-Bratzler shear device (25 kg load cell) and a crosshead speed of 200 mm/min. Down stroke distance was 3 cm (the probe should cut the meat completely). The parameter recorded was the maximum resistance of the sample to shearing or the maximum shear force that is the highest peak (recorded in kg) of the curve. Each core was assessed 2 times, and the twelve peak shear forces recorded per sub-sample were averaged.

## 2.4. Statistical analyses

A simple descriptive Statistics analysis was performed for live, carcass and meat characteristics. A preliminary statistical analysis was performed by calculating the Pearson's correlations between the variables of meat quality (pH, colour, WBSF, PL and CL). Traits with only one observation per animal (final weight, slaughter weight and carcass traits: dressing-out, conformation, fatness, pH, temperature and instrumental color) were analyzed with a GLM with the pre-slaughter treatment (stressed or unstressed) and breed (CH, LI and RE) and their interactions as fixed effects and kill group as random block effect. As all meat quality measurements were taken thrice (1, 7 and 21 days of ageing), the measurement of the dependent variable is repeated. Therefore, traits with 3 observations per animal (meat quality: instrumental color, WBSF, PL and CL) were analyzed using the MIXED procedure with a within-subject (animal) design for repeated measures with unequal spacing as suggested by Littell, Henry, and Ammerman (1998). The model included the pre-slaughter treatment, breed and ageing time

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