# Effect of enriched housing on welfare, production performance and meat quality in finishing lambs: The use of feeder ramps

L.A. Aguayo-Ulloa <sup>a</sup>, G.C. Miranda-de la Lama <sup>b</sup>, M. Pascual-Alonso <sup>a</sup>, J.L. Olleta <sup>a</sup>, M. Villarroel <sup>c</sup>, C. Sañudo<sup>a</sup>, G.A.María<sup>a</sup>.

#### Abstract

This study analyses the effect of environmental enrichment on the welfare, productive traits and meat quality of lambs housed in feedlots. Sixty lambs were placed in enriched (EE) or conventional (CO) pens (3 pens for each treatment, 10 lambs/pen) where EE had a wooden platform with ramps that provided access to a concentrate hopper, cereal straw as bedding and forage, and one play ramp. The CO pen was barren, similar to commercial feedlots. The physiological adaptation response of EE lambs was more efficient than CO, since the latter mobilised more body reserves (i.e., increased NEFA, P < 0.05), and had lower levels of immunity (i.e., increased N/L, P < 0.05), which indicate chronic stress, probably associated with the barren environment. The EE lambs had a higher (P < 0.05) average daily gain, with heavier carcasses and higher fattening scores, as well as lower pHult, higher L\* and b\* values, and lower values of texture (P < 0.05).

Keywords: Full environmental enrichment Finishing lambs; Meat quality; Animal welfare; Productive performance

## 1. Introduction

In the future, one of the main challenges of the sheep industry will be to increase production to satisfy the global demand for meat con-sumption, as one more source of protein in the human diet (Montossi et al., 2013). Along those lines, in Mediterranean countries there has been a trend to adjust traditional pastoral sheep production to more intensive schemes (Bernués, Ruiz, Olaizola, Villarba, & Casasús, 2011; de Rancourt, Fois, Lavín, Tchakérian, & Vallenard, 2006). The development of intensive indoor management programmes, such as lamb feed-lotting, externalises the final stage of fattening to off-farm units (Aguayo-Ulloa et al., 2013). That process stratifies the system in two specialised parts, breeding the flock on the farm (under the farmer's responsibility) and fattening on feedlots, also called classification centres (CCs). These changes in lamb meat production simplify the finishing process for the farmer and improve carcass homogeneity (Miranda-de la Lama, Villarroel, Liste, Escós, & María, 2010; Miranda-de la Lama et al., 2010). However, both farmers and animals have to deal with new problems such as external resource dependency, multiple live transports, social mixing, novel and barren environments, and frequent handling (Aguayo-Ulloa et al., 2013; Miranda-de la Lama, Villarroel, & Maria, 2012). The lack of stimulation leads to boredom, which may cause the development of stereotypies, abnormal behaviour, frustration and stress (Fraser, 1980; Lawrence & Rushen, 1993; Wood-Gush & Beilhartz, 1983).

The new intensive sheep production described can have negative effects on animal welfare and the quality of products delivered to consumers. The welfare of farm animals is a growing public concern and considered a priority for an increasing number of Europeans (European Commission, 2006; Vanhonacker, Verbeke, Van Poucke, & Tuyttens, 2008). New regulations have been developed to improve the production chain of the intensive livestock industry to satisfy consumer demands (European Commission, 2006; Winter, Fry, & Carruthers, 1998).

 <sup>&</sup>lt;sup>a</sup> Department of Animal Production and Food Science, Faculty of Veterinary Science, University of Zaragoza, Spain
<sup>b</sup> Group of Animal Welfare and Sustainable Production, Department of Food Science, Metropolitan Autonomous University, UAM-Lerma, State of Mexico, Mexico

<sup>&</sup>lt;sup>c</sup> Department of Animal Science, E.T.S.I.A. Polytechnic University of Madrid, Spain

Adequate environmental enrichment could reduce negative emo-tional states such as fear and stress associated with the adaptation to a novel environment (i.e., the CC). This will reduce the frustration that animals may experience when they are unable to express their behavioural needs (Hughes & Duncan, 1988; Nicol, 1992; Wood-Gush & Vestergaard, 1989). Environmental modifications can also improve physical health by promoting a wide range of movements to promote skeletal muscle and cardiovascular fitness (Chamove, 1989; Fraser, Phillips, & Thompson, 1986; Klont et al., 2001). Many ways to improve animal welfare through environmental enrichment have been explored, mainly in pigs (Bracke et al., 2006; van de Weerd & Day, 2009; Vanheukelom, Driessen, & Geers, 2012). The literature review by de Azevedo, Cipreste, and Young (2007) reports that food and structural enrichment are among the most successful. However, in farm animals most enrichment studies concentrate on structural applications and not on food presentation. One alternative in lambs is enrichment using several items (Abou-Ismail, 2011) but no studies have been performed during the fattening period in feedlots. The goal of the present study is to analyse the effect of functional full enrichment on a combined set of variables. Our study is based on the hypothesis that enrichment during the finishing period of fattening may improve the adaptation process of the lambs to a novel environment at the CC, which may optimise welfare and productive performance, thereby preserving farmer income. The effect of the enrichment items (feeder ramps and cereal straw) was measured on physiological welfare indicators, performance productive traits and meat quality variables during the finishing phase of fattening lambs.

### 2. Materials and methods

The study was carried out using the installations of the Animal Experimentation Service of the University of Zaragoza in the Autono-mous Community of Aragon, Spain (41°41′N). All the lambs were raised, transported and slaughtered according to current regulations of the European Commission (1986) for Scientific Procedure Establishments. Experimental protocols were approved by the Animal Experimentation Ethics Committee of the University of Zaragoza (ES 50 297 0012 006).

## 2.1. Study description

A total of 60 healthy Rasa Aragonesa male lambs (65 days old) with an average live weight of 17.13 (±0.18) kg, were allocated into two treatments (weights were balanced across treatments) according to their pen environment during the finishing phase of fattening, which lasted five weeks. Lambs were housed indoors in six pens with 10 lambs each (2.9 × 3.3 m, animal density of 0.95 m<sup>2</sup> per lamb) and three replicates per treatment. The lambs from the enriched environ-ment (EE) were maintained in pens with a wooden platform with ramps that provided access to a concentrate hopper (Fig. 1). The plat-form dimensions were 2.35 m long, 1.55 m wide and 0.35 m high (1.67 m<sup>2</sup>). The ramp slope was approximately 20°. The platform in the pen was attached to the solid fence that separated each pen, allowing the lambs to feed, explore or rest lying down. The lambs from EE were provided with cereal straw as bedding on the floor and as forage in a fodder rake. Additionally a small ramp (0.90 m long, 0.50 m wide, 0.35 m high and 0.08 m2 surface) was situated near the opposite solid fence, but away from the food hopper and fodder rake, to allow lambs to play. The lambs in the conventional environment or controls (CO) were maintained in pens of the same size as EE, but without cereal straw (as bedding or forage) or ramps (common environmental condi-tions in CCs). For hygienic reasons a thin layer of sawdust was added at the beginning of the experiment. All lambs were fed with commercial concentrate (Ovirum Alta Energía®) containing barley, wheat, calcium carbonate, sodium chloride and a vitamin supplement corrector (18% crude protein and 11.5 MJ metabolisable energy/kg DM). Feeding and water consumption were ad libitum. In both treatments the concentrate hopper was wide enough to allow all lambs to eat simultaneously. Water was provided using a float drinker installed in a corner of each pen. Feed consumption (concentrate) was registered to estimate the conversion index during the fattening period. Animals were weighed individually at the beginning of the experimental period (W1) and just before slaughter (W2).

# 2.2. Physiological welfare indicators

Blood samples were taken by jugular venipuncture with vacuum tubes (before final weighing) to evaluate physiological responses to stress (two 4 ml tubes per animal, with and without anticoagulant, EDTA-K3). Samples were collected by the trained personnel that han-dled the animals and they performed the venipuncture in less than 1 min per lamb, as a necessary precaution to avoid sampling error. Sam-ples were kept on ice for a maximum of 2 h and taken to the laboratory for routine haematological measurements. The EDTA plasma and serum were centrifuged at 3000 rpm for 10 min and aliquots were frozen and kept at -30 °C until analysed. An automatic particle counter (Microcell counter F-800 and auto dilutor AD-260, both Sysmex<sup>TM</sup>) was used to count red blood cells (RBCs) and white blood cells (WBCs) (number per litre), haemoglobin (g/dl) and

haematocrit (%). The leukocyte for-mula was estimated from blood swabs on clean slides. Staining was per-formed by the rapid panoptic method using dyes from Química Clinica Aplicada Inc. (QCA). Using an optic immersion microscope we counted and identified 100 leucocytes per sample (neutrophils, lymphocytes, eosinophils, basophils and monocytes). The neutrophil/lymphocyte ratio (N/L) was used as an indicator of chronic stress (Lawrence & Rushen, 1993). Serum samples were used to determine the concentra-tion of glucose (mg/dl, Ref. Glucose AE2-17), and the activity of creatine kinase (CK) (IU/l) (Ref. CK.NAC AE1-13) using a multianalyser ACE® (clinical chemistry system) and reagents from Alfa Wasserman. Serum concentration of non-esterified fatty acid (NEFA) levels was analysed by a multianalyser ACE® (clinical chemistry system of Alfa Wasserman), with commercial kits (NEFA C Ref. 994-75409 of Wako). The concentration of cortisol was determined from plasma (EDTA-K3) by enzyme immunoassay using an "in home-kit" (validated by Chacón, Garcia-Belenguer, Illera del Portal, & Palacio, 2004). Each sample was determined in duplicate from 50 µl of plasma and the results were expressed in nmol/l, with the corresponding controls. Variation coefficients of the analysis, inter- and intra-assay, were 7 and 8% respectively. The concentration of lactate was determined using a Sigma diagnostic kit (lactate no. 735-10) and spectrophoto-meter (Lambda 5, Perkin Elmer).

Temperatures were taken by infrared thermography (IRT). Lambs were randomly captured and restrained by a trained handler. Restraint continued for 1 min during which a photograph of the left eye was taken with an IR camera (Testo 880 thermal imaging camera; Testo AG, Lenzkirch, Germany) to evaluate acute stress response produced by handling (Stewart et al., 2007). All images were collected from the left side of the lamb (approximate distance of 20 cm). The built-in lens (24°) was used and the camera was calibrated for the current room temperature and relative humidity. The emissivity value used was 0.98, which is recommended by the camera manufacturer for biological tissues. A clear infrared image (precise location and perfect focus) was chosen from the set of pictures taken of each animal. Image analysis software (IRSoft™ software, Testo AG, Lenzkirch, Germany) was used to determine the maximum temperature within an oval area traced around the eye, including the eyeball and approximately 1 cm around the outside of the eyelids (Stewart et al., 2007).

## 2.3. Performance and meat quality variables

The amount of concentrate added to the feeder and feed remains (at the end of experiment) was recorded. We estimated the total consump-tion of concentrate (TCC) as the difference between concentrate added and concentrate remaining in the feeder. Average daily gain (ADG) was estimated by the difference between W1 – W2 (WG) divided by the total fattening period (34 days). The concentrate conversion index (CCI) was estimated as TCC/WG. The animals were slaughtered within the weight range of the Ternasco-type category (Sañudo, Santolaria, María, Osorio, & Sierra, 1996; Sañudo et al., 1998) at an EU-approved abattoir located in the city of Zaragoza.

After slaughter, carcasses were stored in cold rooms at 2 °C for 24 h. Cold carcasses were weighed (CW) at 24 h (at 1–2 °C) in the cold room. The extent of bruising on the carcasses was estimated visually using an adapted scale from Miranda-de la Lama et al. (2009) with a score of 0 (no bruising), 1 (slight bruising), 2 (moderate bruising) or 3 (high bruising). Carcass conformation score (CS) and carcass fatness (FS) were graded according to the European Classification System (European Union, 1993), the EUROP conformation scale (converted to a 15 point scale) and the carcass fatness scale (converted to a 15 point scale). After chilling for 24 h the left rack was removed from the T1 to L6 vertebrae. The pH at 24 h (pHult) of the M. longissimus was assessed using a portable pH metre (fitted with a penetration electrode 52-00 from Crison), which was inserted into a small incision in the left loin (L2–L3 vertebrae). The pH metre was re-calibrated after every five sam-ples, using two standard buffer solutions at pH 7.02 and 4.00. After that, the left rack was transferred to the Meat Laboratory at the Faculty of Veterinary Medicine of the University of Zaragoza without disrupting the cold chain.

The M. longissimus was removed from the rack to prepare the sam-ples. A section of meat from T10 to T13 vertebrae was weighed (FMW)(mean 145 g), vacuum-packed, frozen, and stored at -20 °C after 72 h of ageing, to evaluate thawing loss (TL%) and cooking loss (CL%) and to perform the Warner–Bratzler test on. Samples were thawed in their vacuum-sealed plastic bags for 24 h in a refrigerator (2–4 °C) before testing. The thawed samples were then weighed (TMW) (mean 137 g) and cooked for 35 min in plastic bags at 75 °C in a water bath (GLF-D3006), until the internal temperature of the meat (measured with a penetration thermometer) reached 70 °C, then cooled for 30 min under flowing cold water. After the samples were cooled to room temperature, they were blotted dry using paper towels and weighed (CMW). The TL% and CL% were calculated as [TL% = 100 – (TMW × 100) / FMW] and [CL% = 100 – (CMW × 100) / TMW]. The texture of the cooked meat was measured with a Warner–Bratzler device, using an Instron 4301 equipped with a Warner–Bratzler shear. To cut 1cm2 pieces (in the direction of

the muscle fibres), we used a digital cal-ibre Mitutoyo series 500 (Mitutoyo Corporation, Aurora, IL, USA). For each animal, three measurements were taken. Shear force (kg/cm2), maximum stress (kg/cm2), and toughness (kg) were measured as de-scribed by Campo et al. (2000). The gauge and the gauge length of the sample were 10 mm and 30 mm, respectively. Samples were sheared perpendicularly to the grain. The load cell was 100 kg (minimum load level of 0.001 kg), the crosshead speed was 150 mm/min (high exten-sion limit = 30 mm), and the sampling rate was 20 points/s. Colour was estimated at 24 h post-mortem and after 15 min of blooming using a Minolta CM 200 calibrated chromameter with a standard illumi-nant D65 and a  $10^{\circ}$  observer with an aperture size of 2.54 cm, following the CIE L\*a\*b\* system, to measure the colour of fresh meat on the cut surface of the T13 vertebra of the M. longissimus. Chroma (C\*) and hue (H\*) indices were calculated as C\* = (a\*2 +b\*2)0.5 (related to the quantity of pigment) and hue as H\* = 1 / tan (b\*/a\*) (the attribute of a colour perception). Final values were the average of three measurements.

## 2.4. Statistical analysis

Production and meat quality data were analysed using least squares methods of the GLM procedure of SAS (SAS, 1988), fitting a one-way model with a fixed effect of environmental enrichment (two levels). The general representation of the model used was: y = Xb + e, where y = Xb + e original full model included the fixed effect in the model with the association matrix x = Xb + e, where y = Xb + e original full model included the effect of the replicate (fitting animals nested within pens), which was found to be non-significant and consequently was dropped from the model. Meat and carcass quality variables were co-varied with cold carcass weight. A probability of y = xb + e or y = xb + e

# 3. Results

In general, the physiological adaptation response to fattening was more efficient in EE lambs (ramps and straw) compared to the conventional environment (barren). The EE lambs also had improved productive performance and meat quality. No clinical health problems or injuries were observed throughout the trial in the lambs from either treatment.

## 3.1. Physiological variables

The least squares means ( $\pm$ SE) for plasmatic and haematological stress indicators are shown in Table 1. Glucose levels were significantly (P b 0.05) higher (16.3%) in EE lambs but NEFA levels were significantly (P b 0.05) higher (133%) in CO lambs. No significant differences between treatments were detected for cortisol, lactate or CK. The N/L ratio was significantly (P b 0.05) higher (42.1%) in CO lambs. No significant differences were found between treatments for the other haema-tological variables. The IRT values were similar in both treatments.

## 3.2. Performance and meat quality variable

The least square means (±SE) for the productive and carcass quality traits are presented in Table 2.Significant differences between treatments (P b 0.05) were observed for ADG, W2, CW, FS and pHult. The ADG of EE lambs was 18.3% higher than CO lambs. Likewise, EE lambs had a higher slaughter weight (4.1%), a higher cold carcass weight (6%) and a higher fattening score (0.5 points). No significant differences were found in carcass conformation or bruising.

The least square means (±SE) for meat quality variables are pre-sented in Table 3. With the exception of redness (a\*) and C\*, all meat quality variables were significantly (P b 0.05) affected by the treatment. The EE lambs had significantly lower values (-1.1%) of pHult than CO. Thawing losses and cooking losses were higher in the meat from EE lambs with a difference of 1.18 (27%) and 0.74 (4.9%) percentage points with respect to CO lambs. Related to meat colour variables, L\* and b\* were higher in the meat from EE lambs (difference of 5% and 12.6%, re-spectively). The H\* values were higher in EE lambs (difference of 9.6%). The texture variables assessed by Warner–Bratzler indicated that shear force, maximum stress and toughness values were lower in the meat from EE lambs (-19.7%, -18.8% and -27.9%, respectively).

#### 4. Discussion

Little is known about the effect of environmental enrichment on the production and meat quality of feedlot lambs but the results obtained in this study suggest that it can help lambs to adjust to indoor feedlots (CCs) by stimulating exercise, foraging and exploration. The positive ef-fects of straw have been demonstrated previously by Teixeira, Miranda-de la Lama, Villarroel, et al. (2012) and Teixeira, Miranda-de la Lama, Pascual-Alonso, et al. (2013) using the same type of lamb. Previous studies have analysed the effect of environmental enrichment on welfare and production, but mainly in pigs (Bracke et al., 2006; van de Weerd & Day, 2009). While the results obtained in those studies are not directly applicable to lamb production, in Spain the finishing phase of fattening lambs is as intensive as that for pigs (Miranda-de la Lama, Villarroel, et al., 2010).

# 4.1. Physiological variables

Lambs fattened in feedlots under intensive conditions tend to present increased HPA activity compared to non-confined systems (Miranda-de la Lama, Rivero, et al., 2010). However, in our study we did not find significant differences in cortisol between treatments, which indicates that lambs coped successfully with the initial acute stressors associated with the fattening environment. Moreover, inten-sive feedlots can be a source of chronic stress, mainly associated with high animal density, poor environment and minimal sensory stimula-tion (Wood-Gush & Vestergaard, 1989). In our study the higher N/L ratio found in CO lambs could indicate chronic stress, but the values were not accompanied by clinical signs of disease.

The differences in glucose and NEFA between treatments could be due to several causes. In general, at rest, the skeletal muscle of sheep uses considerable amounts of blood glucose. As exercise increases, the liver releases additional glucose which is used by the muscle (Maurya, Sejian, Kumar, Singh, & Naqvi, 2012). Pethick (1993) indicated that the rate of glucose release by the liver and uptake by the muscle is sustained and matched during mild exercise (i.e., no hypoglycaemia). That could explain the high levels of plasma glucose in EE lambs, since they had more exercise (going up and down the ramp). Additionally, Klont et al. (2001) note that an increased activity level may result in higher capillary densities in muscle during the rearing period, allowing animals to cope better with stressful events. Considering that these an-imals are very young when they begin the fattening process, moderate exercise could influence the efficiency of their energy use. The higher levels of NEFA found in controls may indicate the presence of underlying multifactorial chronic stressors, including the absence of adequate substrate for behavioural or physiological needs (Hughes & Duncan, 1988). In lambs it has been shown that higher plasma NEFA concentra-tions indicate a breakdown of fat in response to elevated energy demand (Adewuyi, Gruys, & van Eerdenburg, 2005). The higher NEFA and lower plasma glucose in controls indicate a negative energy balance, which is suggestive of chronic stress, possibly linked to a psychological state of depression, as suggested previously by de Jong et al. (2000) and de Groot, de Jong, Prelle, and Koolhaas (2000).

Haematological variables are especially useful in assessing chronic stress related with housing and feeding management (Ramos & Mormède, 1997). A useful measure of the sustained effect of stress is the N/L ratio (Blecha, 2000). As mentioned above, our results show that the N/L ratio was higher in control lambs, indicating immune suppression. Distress can increase the amount of neutrophils and decrease lymphocytes and eosinophils (Kannan et al., 2000; Schaefer, Jones, & Stanley, 1997). Our results agree with Miranda-de la Lama, Rivero, et al. (2010) who reported immune suppression as a conse-quence of the cumulative effect of factors associated with a barren envi-ronment and poor handling at the CC. There were no significant differences in the remaining haematological variables between groups, and their values fell within the normal ranges for this type of animal (Kaneko, Harvey, & Bruss, 1997). Although some physiological stress in-dicators were affected, they did not represent an acute stress response with a high biological cost (Hargreaves & Hutson, 1990). Those findings are substantiated by the lack of a difference between treatments in IRT values.

# 4.2. Performance and meat quality variables

Animal welfare is difficult to measure objectively using simple tech-niques. Itisoften difficult to interpret standard physiological, pro-ductive and behavioural indicators of stress separately (Barnett & Hemsworth, 1990; Mendl, 1991; Rushen, 1991). However, our study provides information to help understand the consequences of stress on the productive performance of lambs and the quality of the product delivered to consumers. Despite the technical difficulties involved in assessing the success of enrichment in livestock animals (Newberry, 1995), productive performance and meat quality analyses provide a practical method to evaluate the economic consequences. Several types of enrichments aimed at enhancing performance have been analysed in other livestock species (Beattie, O'Connell, & Moss, 2000; Enfält et al., 1993; Flint & Murray, 2001; Fraser et al., 1986; Klont et al., 2001). In our study, lambs from the enriched system grew faster and produced improved carcasses, with consequent benefits for meat quality. Similar results were found by Flint and Murray (2001), who found that goats from an enriched feedlot grew nearly twice the rate of goats on a barren feedlot. Pigs given straw as enrichment have higher growth rates with thicker back-fat than pigs finished in a barren envi-ronment (Beattie et al., 2000). Other studies find no difference in the final live-weight between animals given more space and motivated to exercise (Enfält et al., 1993; Fraser et al., 1986) or provided straw bed-ding as enrichment from birth to slaughter (Klont et al., 2001). Similarly, previous studies in lambs have failed to find differences when compar-ing traditional systems of feeding to current industrial systems (Aguayo-Ulloa et al., 2013) or the use of straw for forage and bedding (Teixeira, Miranda-de la Lama, Villarroel, et al., 2012).

The enhanced productive and carcass quality of enriched lambs may be due to a reduction in chronic stress which allows them to use feed more efficiently. That hypothesis is corroborated by the physiological variables, which indicate that the biological cost of adapting to fattening was lower in enriched lambs. Probably, the control lambs in the barren environment were less stimulated and underwent a certain degree of boredom and depression that resulted in redirecting behaviour toward pen mates or decreasing food intake, as seen in pigs (Beattie et al., 2000) or goats (Flint & Murray, 2001). Furthermore, the use of ramps provides repeated daily exercise which lambs may get used to and be able to re-cover energy faster.

Enfält et al. (1993) observed that exercised pigs had shorter car-casses than non-exercised controls. Although we did not measure carcass length, there were no significant differences in carcass confor-mation between treatments. However, EE lambs had higher fatness scores, which could be an advantage in light lambs since fat acts as a protective layer to avoid evaporative losses, which would be greater in leaner meat (Tichenor, 1969). In that way, the EE carcasses could have a higher appreciation on the market as they probably lose less weight during chilling.

Carcass bruises are an extreme (Knowles, 1998) and important indi-cator of the quality of preslaughter handling (Miranda-de la Lama et al., 2009), with commercial and welfare implications. When bruises affect muscle, carcass value can be downgraded, leading to economic losses (Strappini, Frankena, Metz, Gallo, & Kemp, 2012; Taruman & Gallo, 2008). In our study, bruising was very low and not influenced by treat-ment, which indicates good preslaughter handling at the abattoir. The level of bruising was similar to Miranda-de la Lama et al. (2009), and slightly higher than Liste et al. (2011) using the same type of lamb.

The final pH and colour values in EE lambs were related with posi-tive productive performance traits. Tarrant (1989) and then later Apple et al. (1995) suggest that the mechanism of ante-mortem glyco-genolysis in sheep is more likely related to epinephrine release in re-sponse to an emotional stressor than to a contractile mechanism, as occurs in cattle. The lower pHult in enriched lambs suggests that they were less susceptible to stress, had more stored energy and more effi-cient glycogen reposition during preslaughter handling, which coin-cides with Klont et al. (2001) and Essén-Gustavsson et al. (1988). However, the meat pH values observed in both treatments can be con-sidered normal for light lambs (Ripoll, Joy, Muñoz, & Albertí, 2008).

Meat samples from EE lambs lost more water than controls during cooking. That may be explained by the differences in pHult since Bouton, Harris, and Shorthose (1971) found that water losses after cooking decrease linearly with increasing pH. Moreover, decreased cooking losses have been associated with increased water-holding ca-pacity, which is also related to higher pHult. Our results agree with Enfält et al. (1993), who found a greater drip loss in exercised pigs. However, Beattie et al. (2000) and Klont et al. (2001) report that enriched environments may be associated with lower cooking losses, which may be caused by an improvement in water-binding capacity. On the other hand, Teixeira, Miranda-de la Lama, Villarroel, et al.(2012) found no differences between enriched lambs and non-enriched lambs for this trait, reporting lower values than the present study. A positive relationship between cooking losses and fatness has been suggested by Babiker, El khider, and Shafie (1990) and Kemp, Shelley, Ely, and Moody (1972), which agrees with our findings. How-ever, Sañudo, Alfonso, Sánchez, Delfa, and Teixeira (2000) hypothesised that a lack of protective fat in leaner carcasses could also favour losses by facilitating extra alteration in protein structure during chilling and cooking. Other studies report that when fatness differences are small (as in our case), the relationship between fat and losses is insignificant (Dransfield, Nute, Hogg, & Walters, 1990; Kadim, Purchas, Davies, Rae, & Barton, 1993; Sañudo et al., 1996).

Meat colour is one of the most important aspects of meat appear-ance, which consumers use as an indicator of quality and freshness (Faustman & Cassens, 1990). Spanish consumers prefer pale meat from light lambs (Sañudo et al., 2007). Our results indicate that lambs fattened under enriched housing have paler meat with a higher tone than non-enriched lambs, making it more valuable. Meat colour is high-ly related to pH when the former is near 6.0 (Apple et al., 1995), which was not our case. Thus, other factors besides pH may have been involved in affecting meat colour. One factor could be the higher fatness in enriched lambs, as suggested by Kadim et al. (1993) who notes that an-imals selected for greater fat thickness have a tendency to produce lighter-coloured meat and have a lower pH. Sañudo et al. (2000) found no differences in lightness in lambs from different fat classes but some differences in a\* and b\* values. Another factor affecting meat colour is structural change in the muscle protein of enriched lambs that had more exercise, as suggested in Enfält et al. (1993).

Meat tenderness is another highly valuable attribute for consumers of light lambs (Sañudo et al., 2000). Shear force measured by Warner–Bratzler is widely used to evaluate cooked meat and reflects the tough-ness of both myofibrillar and connective tissues. The meat from enriched lambs was tenderer than controls. Differences in texture may be related to exercise, as found by Carrasco, Panea, Ripoll, Sans, and Joy (2009). Moderate exercise has been related with a greater muscle volume and a higher myofibrillar protein to total collagen ratio (Aalhus, Price, Shand, & Hawrysh, 1991), leading to a more tender meat. Another possible explanation could be related with ultimate pH.

When ultimate pH increases from 5.6 to 6.0, tenderness decreases (Bouton et al., 1971; Graafhuis, Honikel, Devine, & Chrystall, 1991). Al-though in our study, meat pH from lambs of both treatments fell below this range, the pH of control lambs was higher and close to 5.6. Therefore, this could be related with proteolytic activity at intermediate pH (and with the activation of calpain proteases (Koohmaraie, 1988)).

# 5. Conclusions

The results obtained in this study indicate that enrichment improves the adaptation process of the lambs to a novel environment with conse-quent benefits for animal well-being and the quality of the product. Lambs from the control group (no enrichment) had physiological indicators compatible with chronic stress and immune suppression, probably related to a low level of sensory stimulation in a barren environment.

We believe that the type of enrichment used in our study can be applied as a tool or animal welfare oriented practise to prevent welfare problems for lambs adapting to a new system. The multiple functional items proposed represent a feasible alternative to enrich the environ-ment in indoor feedlots (CCs), preserving the quality of the meat deliv-ered to consumers. Taking into account that European legislation regarding sheep production is currently being considered in the UE, our results may be valuable to help legislators in the development of scientificbasedregulations.

#### References

- [1] Aalhus, J. L., Price, M.A., Shand, P. J., & Hawrysh, Z. J. (1991). Endurance-exercised growing sheep: II. Tenderness increase and change in meat quality. Meat Science, 29, 57–68.
- [2] Abou-Ismail, U. A. (2011). Are the effects of enrichment due to the presence of multiple items or a particular item in the cages of laboratory rat? Applied Animal Behaviour Science, 134, 72–82.
- [3] Adewuyi, A. A., Gruys, E., & van Eerdenburg, F. J. (2005). Non-esterifed fatty acids (NEFA) in dairy cattle. A review. Veterinary Quarterly, 27, 117–126.
- [4] Aguayo-Ulloa, L. A., Miranda-de la Lama, G. C., Pascual-Alonso, M., Fuchs, K., Olleta, J. L., Campo, M. M., Villarroel, M., & María, G. A. (2013). Effect of feeding regime during finishing on lamb welfare, production performance and meat quality. Small Ruminant Research, 111, 147–156.
- [5] Apple, J. K., Dikeman, M. E., Minton, J. E., McMurphy, R. M., Feed, M. R., Leith, D. E., & Unruch, J. A. (1995). Effects of restraint and isolation stress and epidural blockade on endocrine and blood metabolite status, muscle glycogen metabolism, and inci- dence of dark-cutting longissimus muscle of sheep. Journal of Animal Science, 73, 2295–2307.
- [6] Babiker, S. A., El khider, I. A., & Shafie, S. A. (1990). Chemical composition and quality at-tributes of goat meat and lamb. Meat Science, 28, 273–277.
- [7] Barnett, J. L., & Hemsworth, P. H. (1990). The validity of physiological and behavioural measures of animal welfare. Applied Animal Behaviour Science, 25, 177–187.
- [8] Beattie, V. E., O'Connell, N. E., & Moss, B. W. (2000). Influence of environmental enrichment on the behaviour, performance and meat quality of domestic pigs. Livestock Production Science, 65, 71–79.
- [9] Bernués, A., Ruiz, R., Olaizola, A., Villarba, D., & Casasús, I. (2011). Sustainability of pasture-based livestock farming systems in the European Mediterranean context: Synergies and trade-offs. Livestock Science, 139, 44–57.
- [10] Blecha, F. (2000). Immune system response to stress. In G. P. Moberg, & J. A. Mench (Eds.), The biology of animal stress: Basic principles and implications for animal welfare (pp. 111–122). New York, New York, USA: CABI Publishing.
- [11] Bouton, P. E., Harris, P. V., & Shorthose, W. R. (1971). Effect of ultimate pH upon the water holding capacity and tenderness of mutton. Journal of Food Science, 36, 435–439.
- [12] Bracke, M. B.M., Zonderland, J. J., Lenskens, P., Schouten, W. G. P., Vermeer, P., Spoolder, H. A.M., Hendriks, H. J. M., & Hopster, H. (2006). Formalised review of environmental enrichment for pigs in relation to political decision making. Applied Animal Behaviour Science, 98(3–4), 165–182.
- [13] Campo, M. M., Santolaria, P., Sañudo, C., Lepetit, J., Olleta, J. L., Panea, B., & Albertí, P. (2000). Assessment of breed type and ageing time effects on beef meat quality using two different texture devices. Meat Science, 55, 371–378.
- [14] Carrasco, S., Panea, B., Ripoll, G., Sans, A., & Joy, M. (2009). Influence of feeding systems on cortisol levels, fat colour and instrumental meat quality in light lambs. Meat Science, 83, 50–56.
- [15] Chacón, G., Garcia-Belenguer, S., Illera del Portal, J. C., & Palacio, J. (2004). Validation of an EIA technique for the determination of salivary cortisol in cattle. Spanish Journal of Agricultural Research, 2(1), 45–51.
- [16] Chamove, A. S. (1989). Environmental enrichment: A review. Animal Technology, 40, 155–178.
- [17] de Azevedo, C. S., Cipreste, C. F., & Young, R. J. (2007). Environmental enrichment: A GAP analysis. Applied Animal Behaviour Science, 102(3-4), 329-343.
- [18] de Groot, J., de Jong, I. C., Prelle, I. T., & Koolhaas, J. M. (2000). Immunity in barren and enriched housed pigs differing in baseline cortisol concentration. Physiology & Behavior, 71, 217–223.
- [19] de Jong, I. C., Prelle, I. T., van de Burgwal, J. A., Lambooij, E., Korte, S. M., Blokhuis, H. J., & Koolhass, J. M. (2000). Effects of environmental enrichment on behavioral responses to novelty, learning, and memory, and the circadian rhythm in cortisol in growing pigs. Physiology Behavior, 68, 571–578

- [20] de Rancourt, M., Fois, N., Lavín, M. P., Tchakérian, E., & Vallenard, F. (2006). Mediterranean sheep and goat production: An uncertain future. Small Ruminant Research, 62, 167–179.
- [21] Dransfield, E., Nute, G. R., Hogg, B. W., & Walters, B. R. (1990). Carcass and eating quality of ram, castrated ram and ewe lambs. Animal Production, 50, 291–299.
- [22] EEC lamb carcass classification system regulations, number: 2137/92 and 461/93 (1993). Office for Official Publications of the European Communities, L-2985. Luxembourg.
- [23] Enfält, A.C., Lundström, K., Hansson, I., Karlsson, A., Essén-Gustavsson, B., & Håkansson, J. (1993). Moderate indoor exercise: Effect on production and carcass traits, muscle en-zyme activities and meat quality in pigs. Animal Science, 57(01), 127.
- [24] Essén-Gustavsson, B., Lundström, K., Larsson, G., Lindholm, A., Nordin, A.C., Hansson, I., & Tornberg, E. (1988). The effect during growth of moderate exercise on muscle meta-bolic characteristics in vivo and relation to meat quality and sensory properties. Pro- ceedings of the Thirty-Fourth International Congress of Meat Science And Technology, Brisbane, Australia (pp. 27–30).
- [25] European Commission (1986). Scientific procedure and breeding of animals for use in scientific procedure establishments. Directive 86/609/ECC 1986.
- [26] European Commission (2006). Commission working document on a community action plan on the protection and welfare of animals 2006–2010. Available in: http:// europa.eu.int/comm/food/animal/welfare/work\_doc\_strategic\_basis230106\_en.pdf (Accessed January 20, 2013)
- [27] Faustman, C., & Cassens, R. G. (1990). The biochemical basis for decoloration in fresh meat: A review. Journal of Muscle Foods, 1, 217–243.
- [28] Flint, M., & Murray, P. J. (2001). Lot-fed goat: The advantages of using an enriched envi-ronment. Australian Journal of Experimental Agriculture, 41, 473–476.
- [29] Fraser, A. F. (1980). Ethology, welfare and preventive medicine for livestock (editorial). Applied Animal Ethology, 6, 103–109.
- [30] Fraser, D., Phillips, P. A., & Thompson, B. K. (1986). A test of a free-access two-level pen for fattening pigs. Animal Science, 42, 269.
- [31] Graafhuis, A. E., Honikel, K. O., Devine, C. E., & Chrystall, B. B. (1991). Tenderness of different muscles cooked to different temperatures and assessed by different methods. Proceedings of the thirty-seventh ICoMST (pp. 365–368). Kulmbach: Federal Centre for Meat Research.
- [32] Hargreaves, A. L., & Hutson, G. D. (1990). The stress response in sheep during routine han-dling procedures. Applied Animal Behaviour Science, 26, 83–90.
- [33] Hughes, B. O., & Duncan, I. J. H. (1988). The notion of ethological 'need', models of moti- vation and animal welfare. Animal Behaviour, 36, 1696–1707.
- [34] Kadim, I. T., Purchas, R. W., Davies, A. S., Rae, A. L., & Barton, R. A. (1993). Meat quality and muscle fibre type characteristic of Southdown rams from high and low back fat selection lines. Meat Science, 33, 97–109.
- [35] Kaneko, J. J., Harvey, J. W., & Bruss, M. L. (1997). Appendixes. In Jerry J. Kaneko, John W. Harvey, & Michael L. Bruss (Eds.), Clinical biochemistry of domestic animals (pp. 885–905) (5th ed.). San Diego: Academic PressSan Diego.
- [36] Kannan, G., Terrill, T. H., Kouakou, B., Gazal, O. S., Gealye, S., Amoach, E. A, & Smaké, S. (2000). Transportation of goat: Effects on physiological stress responses and live weight loss. Journal of Animal Science, 78, 1450–1457.
- [37] Kemp, J.D., Shelley, J. M., Ely, D.G., & Moody, W. G. (1972). Effect of castration and slaugh- ter weigh on fatness, cooking losses and palatability of lamb. Journal of Animal Science, 33, 97–109.
- [38] Klont, R. E., Hulsegge, B., Hoving-Bolink, A. H., Gerritzen, M.A., Kurt, E., Winkelman- Goedhart, H. A, de Jong, I. C., & Kranenet, R. W. (2001). Relationships between behav- ioral and meat quality characteristics of pigs raised under barren and enriched hous- ing conditions. Journal of Animal Science, 79, 2835–2843.
- [39] Knowles, T. G. (1998). A review of road transport of slaughter sheep. Veterinary Record, 143, 212–249.
- [40] Koohmaraie, M. (1988). The role of endogenouse proteases in meat tenderness. Reecip. Meat Conference. Proc., 41. (pp. 89), 89.
- [41] Lawrence, A.B., & Rushen, J. (1993). Stereotypic animal behaviour: Fundamentals and applications to welfare. UK: CABI.
- [42] Liste, G., Miranda-de la Lama, G. C., Campo, M. M., Villarroel, M., Muela, E., & María, G. A. (2011). Effect of lairage on lamb welfare and meat quality. Animal Production Science, 51, 1–7.

- [43] Maurya, V. P., Sejian, V., Kumar, K., Singh, G., & Naqvi, S. M. K. (2012). Walking stress in-fluence on livestock production. In V. Sejian (Eds.), Environmental stress and ameliora- tion in livestock production (pp. 75–95). Berlin Heidelberg: Springer-Verlag (Chapter 4).
- [44] Mendl, M. (1991). Some problems with the concept of a cut-off point for determining when an animal's welfare is at risk. Applied Animal Behaviour Science, 31, 139–146.
- [45] Miranda-de la Lama, G. C., Rivero, L., Chacón, G., Garcia-Belenguer, S., Villarroel, M., & Maria, G. A. (2010). Effect of the pre-slaughter logistic chain on some indicators of welfare in lambs. Livestock Science, 128, 52–59.
- [46] Miranda-de la Lama, G. C., Villarroel, M., Liste, G., Escós, J., & María, G. A. (2010). Critical points in the pre-slaughter logistic chain of lambs in Spain that may compromise the animal's welfare. Small Ruminant Research, 90, 174–178.
- [47] Miranda-de la Lama, G. C., Villarroel, M., & Maria, G. A. (2012). Behavioural and physiolog- ical profiles following exposure to novel environment and social mixing in lambs. Small Ruminant Research, 103, 158–163.
- [48] Miranda-de la Lama, G. C., Villarroel, M., Olleta, J. L., Alierta, S., Sañudo, C., & Maria, G. A. (2009). Effect of the pre-slaughter logistic chain on meat quality of lambs. Meat Science, 83, 604 609.
- [49] Montossi, F., Font, I., Furnols, M., del Campo, M., San Julián, R., Brito, G., & Sañudo, C. (2013). Sustainable sheep production and consumer preferences trend: Compatibilities, contradictions, and unresolved dilemmas. Meat Science, 5, 772–789.
- [50] Newberry, R. C. (1995). Environmental enrichment: Increasing the biological relevance of
- [51] captive environments. Applied Animal Behaviour Science, 44, 229–243.
- [52] Nicol, C. J. (1992). Effects of environmental enrichment and gentle handling on behaviour and fear responses of transported broilers. Applied Animal Behaviour Science, 33, 367–380.
- [53] Pethick, D. W. (1993). Carbohydrate and lipid oxidation during exercise. Australian Journal of Agriculture Research, 44, 431–441.
- [54] Ramos, A., & Mormède, P. (1997). Stress and emotionality: A multidimensional and ge- netic approach. Neuroscience and Biobehavioral Reviews, 22, 33–57.
- [55] Ripoll, G., Joy, M., Muñoz, F., & Albertí, P. (2008). Meat and fat color as a tool to trace grass feeding systems in light lamb production. Meat Science, 80, 239–248.
- [56] Rushen, J. (1991). Problems associated with the interpretation of physiological data in the assessment of animal welfare. Applied Animal Behaviour Science, 28, 381–386.
- [57] Sañudo, C., Santolaria, M. P., María, G., Osorio, M., & Sierra, I. (1996). Influence of carcass weight on instrumental and sensory lamb meat quality in intensive production systems. Meat Science, 42, 195–202.
- [58] Sañudo, C., Sanchez, A., & Alfonso, M. (1998). Small ruminant production systems and factors affecting lamb meat quality. Meat Science, 49, S29–S64.
- [59] Sañudo, C., Alfonso, M., Sánchez, A., Delfa, R., & Teixeira, A. (2000). Carcass and meat qual- ity in light lambs from different fat classes in the EU carcass classification system. Meat Science, 56, 89–94.