# Influence of crude glycerin inclusion in the diet, genetic group and postmortem aging time on color of three bovine muscles

Oliveira, I.M.<sup>1</sup>, \*<u>Paulino, P.V.R.<sup>1</sup></u>, Monnerat, J.P.I.S.<sup>1</sup>, Serão, N.V.L.<sup>2</sup>, Couto, V.R.M.<sup>1</sup>, Duarte, M.S.<sup>1</sup>, Mezzomo, R.<sup>1</sup>, Silva, L.H.P.<sup>1</sup>, Moura, L.S.<sup>1</sup>, Teixeira, C.R.V<sup>1</sup>.

> <sup>1</sup>Universidade Federal de Viçosa, Department of Animal Science, Viçosa, Brazil. INCT-CA - CNPq <sup>2</sup>University of Illinois, Department of Animal Science, Urbana, United States

Abstract— The study objective was to evaluate the effects of crude glycerin (CG) inclusion in the diet, genetic group and postmortem aging on color of three bovine muscles. Young beef bulls were assigned to a completely randomized design in a 2 x 2 factorial, with repeated measurements (0 and 7 days of aging), two genetic groups (12 Nellore, NE, and 12 F<sub>1</sub> Angus x Nellore, AN, bulls), and two levels of CG inclusion in the diet (5 and 15 %, total DM). Samples of Longissimus dorsi (LD), Gluteus medius (GM) and Biceps femoris (BF), were taken for instrumental color analysis. The surface color measurement (CIE L\* a\* b\*) for each muscle sample was evaluated at d 0 and d 7. There was no interaction (P>0.05) of CG inclusion in the diet x genetic group x aging time on any muscle color. Inclusion of CG in the diet did not affect (P>0.05) color of the muscles evaluated. Aged muscles had increased (P<0.05) b\* and hue angle values, as well as decreased (P<0.05) saturation index (SI) values than non-aged muscles. BF muscle from Nellore bulls had greater (P<0.05) L\* value than of AN. Non-aged BF had greater (P<0.05) L\* value than aged BF, whereas the opposite was observed on LD. Aging for 7 days increased (P<0.05) LD and GM (P<0.05) a\* values. Aging seems to have distinct effects on color of different muscles from Bos indicus cattle carcass, suggesting more studies are needed to unravel color dynamics on beef produced by Zebu cattle.

Keywords – aging time, crude glycerin, muscle color

## **I.** INTRODUCTION

Recently efforts have been made in Brazilian meat industry in order to increase beef production. In addition, with the aim to attend the market demand for a better beef quality, researchers have been seeking for methods that would lead to a better quality of beef, such as the use of biodiesel by-products (i.e. crude glycerin) and the use of crossbred animals.

Technologies such as postmortem aging of meat are also used in order to improve beef quality. However, the use of processing technologies requires studies to evaluate the modifications on the main sensorial aspects of beef such as color.

Meat color has been reported as the main characteristic evaluated by the consumer at the moment of purchase. Changes on meat color are unavoidable and usually indicate bad quality of postmortem handling. Nonetheless, the maintenance and stability of meat color should be investigated aiming to increase its shelf life.

It has been reported that differences among muscles account for most of the differences on stability of meat color. Therefore, recent research evaluating color stability of beef has been focused on pre-mortem and postmortem handling as well as the comparison of different muscles [1].

The objective of the study was to evaluate the effects of crude glycerin (CG) inclusion in the diet, genetic group and postmortem aging on color of three bovine muscles.

# **II.** MATERIALS AND METHODS

This study was conducted at Universidade Federal de Viçosa, Brazil, with 24 bulls, being 12 Nellore (NE) and 12  $\frac{1}{2}$  Angus x  $\frac{1}{2}$  Nellore (AN). The average age and initial body weight were 18 months and 320 kg.

The animals were confined to individual stalls with feeders and drinkers. Diets were formulated to be

isonitrogenous with 13.3% of crude protein (DM basis). The average composition of the experimental diets is presented in Table 1.

Tabela 1. Average composition of the experimental diets

Item (%)	Level of crude glycerin (% total DM)			
	5	15		
Corn silage	50.00	50.00		
Soybean meal	7.15	7.15		
Finely ground corn	34.80	23.10		
Mineral mixture	0.25	0.30		
Urea/ammonium sulphate 9:1	1.0	1.0		
Crude glycerin	5.0	15.0		
Corn gluten meal	0.90	2.55		
Limestone	0.65	0.60		

A completely randomized designed was used in a 2 x 2 factorial arrangement, with two genetic types (NE and AN) and two levels of crude glycerin (5% and 15% of DM, replacing corn).

At the end of the experiment, all animals were slaughtered. All of the slaughters occurred after a 16-h fast and were performed according to the Normative Instruction  $n^0.3$  of 01/13/2000 (Technical Regulation of Methods for Humane Slaughtering of Livestock).

After slaughter, each carcass was divided into identical longitudinal halves and chilled at 4°C for 24 h. After 24 h postmortem chill, muscles samples of *Longissimus dorsi* (LD), *Gluteus medius* (GM) and *Biceps femoris* (BF) were taken from both halves of the carcass of each animal.

Objective color was measured on muscle samples from the right half of the carcass immediately after sampling (day 0). Muscle samples from the left half of the carcass were vacuum packaged and aged for 7 days at 4°C. After postmortem aging period, objective color was measured on aged muscle samples.

Objective color measurement was performed after 40 minutes of air exposition of muscle samples at room temperature, by using a portable colorimeter Minolta CR-400. The CIE  $L^* a^* b^*$  values obtained

by 5 scans were averaged for each muscle sample. Values of saturation index (SI) and hue angle (HA) were calculated as described by [2], using the follow equations: SI =  $((a^*) \times 2 + (b^*) \times 2) \times 0.5$ ; and HA = arctan  $(b^*/a^*)$ .

All statistical procedures were carried out using SAS 9.2 (Statistical Analysis System Institute, Inc., Cary, NC, USA). The data for L\*, a\*, b\*, HA and SI was analyzed as repeated measures following the model:

 $Y_{ijkl} = + D_i + G_j + (DG_i)_j + e_l + M_k + (DM)_{ik} + (GM)_{ik} + (DGM)_{iik} + e_{iikl}$ 

Where:

 $Y_{ijkl}$  is the measured response variable;  $\mu$  is the overall mean valeu;  $D_i$  is the fixed effect of the i<sup>th</sup> level of CG in the diet;  $G_j$  is the fixed effect of the j<sup>th</sup> genetic group;  $(DG)_{ij}$  is the fixed effect of the interaction between the levels of D and G;  $e_l$  is the random error associated with  $Y_{ij}$ , *NID*  $(0, \sigma^2_{el})$ ;  $M_k$  is the fixed effect of the interaction between the levels of ageing;  $(DM)_{ik}$  is the fixed effect of the interaction between the levels of D and M;  $(GM)_{jk}$  is the fixed effect of the interaction between the levels of D and M;  $(DGM)_{ijk}$  is the fixed effect of the interaction between the levels of D, G and M;  $e_{ijkl}$  is the random error associated with  $Y_{iklj}$ , *ND* (0, CS).

Values of instrumental color were repeated in meat sample before and after maturation. The compound symmetry (CS) variance-covariance structure for the repeated measures was used in the analysis. Effects were estimated through restricted maximum likelihood (REML). When first or second order interaction effects were significant (P<0.05), the Tukey-Kramer multiple comparison method was used to test differences among the levels.

## **III.** RESULTS

There was no interaction (P>0.05) between the inclusion of crude glycerin, genetic group and postmortem aging for any color measurements taken.

There was no effect (P>0.05) of crude glycerin on color of the muscles evaluated (Table 2).

Regarding genetic group, NE animals had greater values (P<0.05) of  $L^*$  on BF muscle compared to AN animals (Table 2).

Aged muscles presented greater values of  $L^* b^*$  and HA (P<0.05) than non aged muscles (Table 2).

Tabela 2. Values of instrumental color of *Biceps femoris* muscle

Item	$L^*$	$a^*$	$b^*$	HA	SI
Level of crude					
glycerin					
5%	38.85	18.79	10.70	1.58	29.49
15%	37.80	18.53	10.19	1.65	28.72
P - Value	0.0968	0.6182	0.2972	0.1586	0.4373
Genetic group					
NA	37.39	18.45	10.30	1.62	28.75
NE	39.26	18.87	10.59	1.61	29.46
P - Value	0.0056	0.4335	0.5418	0.9424	0.4730
Postmortem					
aging					
D 0	39.01	18.06	9.67	1.69	27.73
D 7	37.64	19.26	11.25	1.55	30.48
P - Value	0.0081	0 6474	<0.0001	0.0018	<0.0001

Aged GM had greater values (P<0.05) of  $a^*$ ,  $b^*$  and SI (Table 3) than non-aged samples. Values of HA decreased (P<0.05) on aged GM muscle (Table 3).

Tabela 3. Values of instrumental color of *Gluteus Medius* muscle

Item	L*	a*	b*	HUE	IS
Level of crude glycerin					
5%	41.67	20.18	13.11	1.33	33.29
15%	40.68	20.65	12.88	1.46	33.53
P- Value	0.2658	0.5947	0.7527	0.2018	0.8773
Genetic group					
AN	41.37	20.95	13.37	1.37	34.32
NE	40.98	19.88	12.62	1.43	32.50
P- Value	0.6543	0.2294	0.3074	0.5543	0.2487
Postmortem aging					
D 0	40.76	19.67	11.88	1.52	31.55
D 7	41.60	21.16	14.12	1.28	35.27
P - Value	0.1946	0.0085	0.0001	0.0271	0.0006

Aged LD muscle had greater values of  $L^* a^* b^*$  and SI (P<0.05) than non aged samples. There was a reduction (P<0.05) of HA on aged LD muscles (Table 4).

Fabela	4.	Values	of	instrumental	color	of	Longissimus
D <i>orsi</i> n	nusc	cle					

Item	$L^*$	<i>a</i> *	$b^*$	HA	SI
Level of crude glycerin					
5%	39.90	17.30	9.79	1.59	27.08
15%	39.92	17.63	9.72	1.65	27.35
P- Value	0.2150	0.4327	0.8428	0.2846	0.7171
Genetic group					
AN	39.01	17.26	9.66	1.61	26.92
NE	39.81	17.68	9.84	1.63	27.51
P- Value	0.3062	0.3308	0.6158	0.7674	0.4212
Posmortem aging					
D 0	39.07	17.13	8.93	1.75	26.05
D 7	39.75	17.80	10.53	1.49	28.38
P - Value	0.0458	0.0195	< 0.0001	< 0.0001	< 0.0001

## **IV. DISCUSSION**

NE cattle had greater values of  $L^*$  on BF muscle than AN cattle. Genetic type and pre-slaughter handling are two factors that can affect muscle color. In addition, conditions of chilling, such as the space between carcasses in the cooler and carcass fat thickness can directly affect muscle as well [1]. The variation on muscle color among samples aged for 1 and 7 days might be related to the capacity of each individual muscle to resist the change on color that occurs with time, which would be related do some factors, like muscle fiber frequency on different muscles [3].

Postmortem aging affected lightness in different ways on BF and LD muscles. Luminosity is influenced by the amount of water on the surface of the muscle as a consequence of water holding capacity [4]. Differences between aged muscles may be related to differences on water hold capacity and liquid loss.

Postmortem aging increased a\*-values of GM and LD muscles. According to [5], aged meat, even after the equalization of color, still presents a different gradient when compared to non-aged beef. It occurs due to the presence of Fe (III) myoglobin, which is

originated from the reaction between Fe (II) myoglobin and oxygen. As a result, there is formation of metamyoglobin, which present a darker color.

All muscles had greater values of yellowness  $(b^*)$  after 7 days of postmortem aging. Usually aged muscles present darker and browner color and consequently the yellow intensity is increased as well, which is related to storage and temperature. Higher temperatures accelerate the oxidation rate of pigments, increasing the rate of any oxidant reaction inside the tissue [6]. In addition, the increase of temperature increases the oxygen consumption by the tissue, allowing bacteria growth and lipids oxidation. All these process described above contribute to the discoloration and increase of beef yellowness [6].

Values of SI and HA, which are calculated as a function of  $a^*$  and  $b^*$ -values allow to determine color intensity, saturation and to estimate the real darkness of meat. Generally, the discoloration of meat is related to values of HA and SI [7]. In most cases the alterations observed on color coordinates during postmortem aging process are expected since proteolysis decreases water holding capacity [8] and increase the oxidation of color pigments.

Differences on color stability of different muscles due to postmortem aging process can be attributed to greater oxygen consumption in less stable muscles, due to the presence of enzymes that compete with myoglobin for oxygen. As a result, there is an increase on deoximyoglobin formation, which is more susceptible to oxidation than oxymyoglobin [1].

# **V.** CONCLUSION

Postmortem aging seems to have different effects on color of different muscles of Zebu cattle, suggesting that further studies are needed for a better understanding of color of beef from those animals. Crude glycerin levels in the diet up to 15% do not change values of instrumental color of Zebu beef.

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