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Can pre-slaughter stress blood indicators be used to predict dark cutting in lambs? (#520)

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Introduction

Pre-slaughter stress in lambs triggers the hypothalamic-pituitary-adrenal axis to release blood stress indicators (glucose, lactate, cortisol, creatine kinase, lactate dehydrogenase) as the body tries to maintain homeostasis thus resulting in glycogen reserves being depleted ante-mortem [1]. Low glycogen reserves result in low lactate production during the anaerobic glycogen metabolism at post-mortem. Hence, the normal ultimate pH of 5.5 will not be reached thus producing meat with the dark-cutting condition [2]. DFD meat is characterized by a dark color, lower tenderness, high water holding capacity and reduced shelf-life since it promotes rapid microbial growth making it unacceptable to consumers. The objective of this study was to investigate the relationship between ultimate pH and some blood stress indicators at exsanguination.

Methods

A total of 100 ten-month-old Dorper female lambs were humanely slaughtered in a commercial abattoir. Blood samples were collected from each animal within the 30s during exsanguination after stunning and sticking. The University of Fort Hare ethics committee approved all experimental procedures for the study (UFH Ethical clearance number: MUC371SSTE01). The blood samples for the analysis of glucose and lactate levels were collected using 10.0 mL disposable Becton Dickinson vacutainer tubes treated with fluoride oxalate (grey top) whereas those for determination of lactate dehydrogenase, creatine kinase and heat shock protein 70kDa levels were collected using plasma separating vacutainer tubes (SSTTMII gold top). The blood tubes were centrifuged (Model 5403 Centrifuge, Gatenbay Eppendorf GmbH, Engelsdorp, Germany) at 21°C for 1000 g for 15 min. The plasma was then placed in 1.5-mL Eppendorf tubes and stored at –20°C until analysis to avoid loss of bioactivity. Laboratory analysis of plasma glucose and lactate concentrations was carried out using an Olympus AU400 automated chemistry analyzer (Olympus Optical Co. Ltd, Melville, NY, USA) and the Olympus reagent kit for glucose and lactate (Olympus Cat.No.OSR6193). CK and LDH were analyzed using a commercial kit (Kit re-order #442 635: 200 tests/cartridge). HSPA1A was analyzed using a commercial enzyme-linked immunosorbent assay (ELISA) Kit (MBS0334426). Carcass ultimate pH was measured on the right side of each carcass by inserting the piercing probe in the *longissimus* muscle between the 12th and 13th ribs at 24 h after slaughter using a portable pH meter (Hach HQ11d) which was calibrated in buffers with pH 4.00 and 7.00. The data was classified into three groups according to carcass ultimate pH (low pHu, 5.50 < pH < 5.90, n =23; intermediate pHu, 5.90 ≤ pH < 6.10, n = 45; high pHu, pH ≥ 6.10, n = 32) and blood samples were analysed using the PROC GLM procedure of SAS (version 9.1, 2009).

Results

The results indicated that there were no significant differences on the levels of blood parameters amongst the low, intermediate and high ultimate pH groups (Table 1). These results are in contrast with studies by Chulayo *et al.* [3] and Lu *et al.* [4] who found significant differences in cattle. However, the levels of lactate dehydrogenase, creatine kinase, and HASPA1A levels were higher in the high ultimate pH group than the intermediate and low ultimate pH groups. Furthermore, there were significant negative correlations between pH, lactate, and glucose (Table 2). Stempa *et al.* [2] reported similar results, where correlations between pH, glucose, and lactate were observed. When lambs are exposed to pre-slaughter stress (loading, transportation, and feed withdrawal), activates the sympathoadrenal system, which leads to the release of catecholamine by the adrenal medulla. Thus, glycogen from the liver rapidly breaks down, and the levels of blood glucose and lactate increase which negatively affect the pHu and overall meat quality [5].

Table 1: Means and standard deviations of blood variables in groups with low, intermediate and high ultimate pH

Variable	Low pHu	Intermediate pHu	High pHu	P value
Lactate (mmol/L)	4.14 ± 0.37	4.39 ± 0.27	4.28 ± 0.32	0.86
Glucose (mmol/L)	4.68 ± 0.24	4.20 ± 0.17	4.35 ± 0.21	0.28
Lactate dehydrogenase (U/L)	1484.87 ± 56.74	1475.64 ± 40.57	1545.88 ± 48.11	0.51
Creatine Kinase (U/L)	360.00 ± 51.79	446.49 ± 37.02	462.53 ± 43.91	0.28
HSPA1A (ng/ml)	22.09 ± 7.04	22.97 ± 5.03	27.49 ± 5.97	0.80

HSPA1A – Heat Shock Proteins 70kDa, U/L – Units per liter

Table 2: Correlation coefficients among blood variables at exsanguination

	pH	Lactate	Glucose	LDH	CK	HSPA1A
pH	1					
Lactate	-0.93*	1				
Glucose	-0.62*	0.01	1			
LDH	0.36	0.02	-0.16	1		

Notes

CK	0.12	-0.92***	-0.04	0.01	1	
HSPA1A	0.26	0.48	-0.13	0.00	0.04	1

P < 0.05 *, P <0.005, P <0.001***

Conclusion

The results indicated that measuring plasma creatine kinase, lactate dehydrogenase and HSPA1A levels at exsanguination cannot be used to identify the dark-cutting condition in lambs. However, plasma glucose and lactate can be useful indicators for DFD meat in lambs at exsanguination.

Notes