ASSOCIATION OF GENOMIC MARKERS (SNP) IN THE *CAST* GENE WITH TENDERNESS IN SOUTH AFRICAN PUREBRED BEEF CATTLE

Annie Basson[1,2], Lorinda Frylinck[1*] and Phillip E. Strydom[1]

[1] Agricultural Research Council of South Africa, Animal Production (ARC-AP), Irene 0062, South Africa

[2] Department of Animal and Wildlife Science, University of Pretoria (UP), South Africa

* Corresponding author email: lorinda@arc.agric.za

I. INTRODUCTION

Very few studies have validated genomic marker effects on meat quality in South African (SA) beef cattle [1]. Calpastatin controls the enzymatic activity of µ-calpain, which in turn makes the largest contribution to proteolysis and tenderness of beef [2] and the *cast* gene (BTA7) contains QTL for tenderness. Although widely validated and included in commercial SNP selection tools, 3'-UTR *cast* SNPs were not found to be informative in SA crossbred beef, most likely because SNPs were not polymorphic and essentially fixed for the favourable allele [1]. We therefore determined the effects of *cast* SNPs on physical tenderness, measured as Warner-Bratzler shear force (WBSF) and myofibrillar fragmentation length (MFL) during ageing, with or without electrical stimulation (ES or NS) in 5 pure beef breeds. By testing a higher density of SNPs spread across the *cast* gene in purebred beef breeds, we aim to find causative SNPs for tenderness that can be used as genomic selection (GS) tools.

II. MATERIALS AND METHODS

South African purebred beef cattle (n=166) were feedlot-finished on grain-based diets to a final age of 12 months old, with a carcass classification of A2 or A3. Breeds included Angus (n=27), Charolais (n=34), Brahman (n=35), Bonsmara (n=35) and Nguni (n=35), which are often used in cross-breeding. Carcasses were halved and the right side was electrically stimulated for 120 seconds (ES) at 400 V peak and directly chilled (4°C). The left side was not electrically stimulated (NS) and chilling was delayed (10°C for 6 hours). Samples were collected from longissimus thoracis et lumborum (loin muscle), vacuum packed and aged at 2 ± 1°C for 3, 9, 14 and 20 days post-mortem. Bulls were genotyped using a Bovine-HD SNP BeadChip (Illumina, USA). The WBSF was determined by oven-broiling to 70°C core temperature, cooling to 18°C and shearing using a Universal Instron apparatus (Model 4301, England). The MFL was determined for 100 fragments per sample by Video Image Analysis. Data were analyzed using PLINK 1.90 and Haploview 4.2, with a simple phenotypic association analysis of genotypes with WBSF and MFL, using Bonferroni correction.

III. RESULTS AND DISCUSSION

Six SNPs were informative for tenderness (Table 1) and the predominant associations were found between *cast* genotypes and MFL (Table 2). Individual SNPs explained between 7 - 24% of the variation at all ageing periods, where it seemed that the effects of the *cast* genotype was additive to ES, as correlations with MFL were found in both post-slaughter treatment groups.

Table 1 Informative SNPs in the *cast* gene in South African beef cattle genotyped with the Illumina Bovine-HD SNP BeadChip

Chr	Position	Illumina Name	Variant	R^2 values for MFL	R^2 values for WBSF
7	98510114	BovineHD0700028753	rs136632100	0.087 – 0.236	
7	98511880	BovineHD0700028754	rs134385243	0.073 – 0.096	0.069 – 0.113
7	98537976	BovineHD0700028764	rs109102936	0.084 – 0.158	0.067 – 0.096
7	98540675	BovineHD0700028765	rs133997237	0.066 – 0.156	0.071 – 0.105
7	98541844	BovineHD0700028766	rs135693211	0.066 – 0.107	0.077 – 0.081
7	98547086	BovineHD0700028768	rs110136749	0.066 – 0.107	0.077 – 0.081

Of great interest was the correlation between the *cast* genotypes and early to medium ageing periods between d3 - d14 post-mortem for MFL, and between d9 and d14 post-mortem for WBSF. An increase in the frequency of favourable alleles for tenderness has been shown to decrease the variation in beef

tenderness, while accelerated proteolysis (especially during early ageing) can decrease the ageing period and can have a financial advantage, through earlier marketing of beef [3] [4].

Table 2. Correlations[#] between *cast* genotypes and physical tenderness in South African purebred beef cattle

	Ageing	Treatment	–28753*	–28754	–28764	–28765	–28766	–28768
MFL	d3	NS		8.2% (P≤0.0212)	11.5% (P≤0.0009)	13.5% (P≤0.0001)	6.6% (P≤0.0967)	6.6% (P≤0.0966)
		ES		7.3% (P≤0.0509)	10.3% (P≤0.0030)	12.1% (P≤0.0005)	7.5% (P≤0.0412)	7.5% (P≤0.0412)
	d9	NS	14.7% (P≤0.0001)				7.0% (P≤0.0669)	7.0% (P≤0.0669)
		ES		9.6% (P≤0.0058)	15.8% (P≤0.0001)	15.6% (P≤0.0001)	7.2% (P≤0.0531)	7.2% (P≤0.0531)
	d14	NS	23.6% (P≤0.0001)		8.7% (P≤0.0132)	15.4% (P≤0.0001)	10.7% (P≤0.0019)	10.7% (P≤0.0019)
		ES	13.9% (P≤0.0001)	8.7% (P≤0.0127)	8.4% (P≤0.0166)	12.7% (P≤0.0003)		
	d20	NS	8.7% (P≤0.0132)	8.0% (P≤0.0242)	9.2% (P≤0.0080)	11.7% (P≤0.0007)	7.3% (P≤0.0458)	7.4%% (P≤0.0458)
		ES	8.9% (P≤0.0107)			6.6% (P≤0.0959)		
WBSF	d3	NS						
		ES		6.9% (P≤0.0720)				
	d9	NS			3.5% (P≤0.0867)			
		ES		11.3% (P≤0.0011)	9.6% (P≤0.0057)	10.5% (P≤0.0022)	7.7% (P≤0.0330)	7.7% (P≤0.0330)
	d14	NS						
		ES				7.1% (P≤0.0605)	8.1% (P≤0.0229)	8.1% (P≤0.0229)
	d20	NS						
		ES						

* BovineHD07000–

[#] Percentage phenotypic variation explained by genotypes of the *cast* SNPs (R^2 expressed as a percentage)

IV. CONCLUSION

Several SNPs in the *cast* gene were validated for SA beef cattle, where advancement of the "tender" genotypes through GS could result in increased ageing rate, requiring shorter periods of tenderization that could have large cost-sparing effects.

ACKNOWLEDGEMENTS

We acknowledge the ARC-AP for facilities and animal care. The abattoir, histology and biochemistry personnel of Meat Science and Technology for sampling and analyses. This research was supported by a RMRDSA grant.

REFERENCES

1. Frylinck, L., van Wyk, G.L., Smith, T.P.L., Strydom, P.E., van Marle-Köster, E., Webb, E.C., Koohmaraie, M. & Smith, M.F. (2009). Evaluation of biochemical parameters and genetic markers for association with meat tenderness in South African feedlot cattle. Meat Science 83: 657-665.
2. Koohmaraie, M. & Geesink, G.H. (2006). Contribution of postmortem muscle biochemistry to the delivery of consistent meat quality with particular focus on the calpain system. Meat Science 74: 34-43.
3. Gruber, S.L., Tatum, J.D., Engle, T.E., Chapman, P.L., Enns, R.M., Belk, K.E. & Smith, G.C. (2011). Effects of genetic markers and implant strategy on longissimus and gluteus muscle tenderness of calf-fed steers and heifers. Journal of Animal Science 89: 1401-1411.
4. Schenkel, F.S., Miller, S.P., Jiang, Z., Mandell, I.B., Ye, X., Li, H. & Wilton, J.W. (2006). Association of a single nucleotide polymorphism in the calpastatin gene with carcass and meat quality traits of beef cattle. Journal of Animal Science 84: 291-299.