HOSTED BY

ELSEVIER

Contents lists available at ScienceDirect

Electronic Journal of Biotechnology



Short communication

Meat tenderness genetic polymorphisms occurrence and distribution in five Zebu breeds in Mexico



Gaspar Manuel Parra-Bracamonte ^{a,*}, Juan C. Martínez-González ^b, Ana M. Sifuentes-Rincón ^a, Victor R. Moreno-Medina ^a, Eligio Ortega-Rivas ^c

- a Centro de Biotecnología Genómica, Instituto Politécnico Nacional, Boulevard del Maestro SN. Esq. Elías Piña Col. Narciso Mendoza, C.P. 88710, Reynosa, Tamaulipas, Mexico
- b Facultad de Ingeniería y Ciencias, Universidad Autónoma de Tamaulipas, Centro Universitario Victoria, C.P. 87000, Victoria, Tamaulipas, Mexico
- ^c Asociación Mexicana de Criadores de Cebú, Naranjo 1006, Aguila, C.P. 89230, Tampico, Tamaulipas, Mexico

ARTICLE INFO

Article history: Received 25 May 2015 Accepted 15 July 2015 Available online 28 August 2015

Keywords: Calpain Calpastatin Frequencies Genotype SNP

ABSTRACT

Background: The Zebu cattle are represented by a diverse group of breeds in México. Traditionally these breeds have been associated with the tough beef characteristic. Validated genetic markers have the potential to be included in marker-assisted selection and management programs in order to improve traits such as beef tenderness. The incidence and distribution of Calpain and Calpastatin polymorphisms strongly associated with beef tenderness were estimated in registered cattle of five Zebu breeds in Mexico.

Results: A low and in some cases null frequency of favorable C allele of CAPN316 was determined in all breeds. Conversely, a more equilibrated frequency in CAPN4751 and CAST loci was observed.

Conclusions: Although the relatively low occurrence of favorable alleles in assessed loci may limit their use in selection programs, genotyping availability might be a practical and comprehensive tool for introgression programs by marker assisted selection and management as to improve meat tenderness of Zebu breeds.

© 2015 Pontificia Universidad Católica de Valparaíso. Production and hosting by Elsevier B.V. All rights reserved.

1. Introduction

The Zebu cattle are the second group of breeds most successful in terms of their worldwide distribution [1]. This cattle subspecies is widely used in tropical and subtropical regions because of their thermoregulatory ability under high temperatures and humidity and their genetic resistance to diseases and parasites [2].

A significant proportion of the cattle raised in tropical and subtropical regions took the advantage of *Bos indicus* breeds adaptability, with the resulting undesirable increase in toughness of the meat, as estimated in crossbred cattle with a 50% or higher Zebu inheritance [3]. Evidence indicates that a proportion greater than 25% of *B. indicus* inheritance may significantly affect meat tenderness [4]. Some studies have indicated that higher values of shear force, as a mechanical estimation of tenderness, in the meat from the Central and South regions of Mexico are related to the most frequent used Zebu-type animals [5].

Meat tenderness is one of the most important economic traits of beef cattle related to consumer acceptance and satisfaction. However, its improvement is limited by the difficulty and cost of measurement. Different strategies have been explored for genetic improvement of

E-mail addresse: gparra@ipn.mx, pabraman@hotmail.com (G.M. Parra-Bracamonte). Peer review under responsibility of Pontificia Universidad Católica de Valparaíso.

this trait. Genetic markers have been extensively assessed, and some markers in Calpain and Calpastatin genes are documented and validated to be highly associated with meat shear force [6,7,8,9, 10,11,12,13], explaining 20% of genetic variation of meat tenderness [8,14].

The objective of the present report is to assess the frequency of alleles of genetic markers associated with meat tenderness in five Zebu breeds of beef cattle in Mexico.

2. Experimental

Hair follicle samples of Brahman (n=358), Gyr (n=41), Guzerat (n=21), Indubrazil (n=25) and Sardo Negro (n=109) purebred animals from different beef cattle farms registered in the Asociación Mexicana de Criadores de Cebú, were collected.

For this assessment, allele frequency of the micro-Calpain (CAPN) 316 (GenBank accession no. AF252504: CAPN316) 4751 (GenBank accession no. AF248054: CAPN4751) [15], and Calpastatin (CAST) T1 (GenBank accession no. AF159246: CAST-T1) loci [16] loci, were analyzed. Genotyping of the single nucleotide polymorphisms (SNP) was performed using the Sequenom MassARRAY® system (iPLEX GOLD; Sequenom, San Diego, CA, USA). Genotypic and allelic frequencies of all loci were estimated. Hardy–Weinberg equilibrium (HWE) was tested using GENEPOP ver. 4.2 [17].

^{*} Corresponding author.

Table 1Genotype and allele frequencies of Calpain and Calpastatin polymorphisms in five Zebu breeds.

Locus		BRH	GYR	GZT	IBZ	SNG
CAPN316		P < 0.001	P < 0.001	P < 0.001	P < 0.001	P = 0.005
	GG	0.97	1.00	1.00	1.00	0.99
	GC	0.01	0.00	0.00	0.00	0.00
	CC	0.02	0.00	0.00	0.00	0.01
	G	0.9 7	1.00	1.00	1.00	0.99
	C	0.03	0.00	0.00	0.00	0.01
CAPN4751		P = 0.710	P = 1.000	P < 0.001	P = 1.000	P = 1.000
	TT	0.84	0.83	0.95	0.92	0.84
	TC	0.15	0.17	0.05	0.08	0.16
	CC	0.01	0.00	0.00	0.00	0.00
	T	0.92	0.91	0.98	0.96	0.92
	C	0.08	0.09	0.02	0.04	0.08
CAST-T1		P = 1.000	P = 1.000	P = 1.000	P = 0.410	P = 0.848
	CC	0.08	0.19	0.05	0.28	0.18
	CT	0.40	0.49	0.33	0.60	0.48
	TT	0.52	0.32	0.62	0.12	0.34
	C	0.28	0.44	0.21	0.58	0.42
	T	0.72	0.56	0.79	0.42	0.58

BRH: Brahman, GYR: Gyr, GZT: Guzerat, IBZ: Indubrazil, SNG: Sardo Negro. *P* values < 0.05 are statistically significant for Hardy Weinberg deviation test.

3. Results and discussion

In Mexico the Zebu breeds are maintained as purebred animals but also as crossbred animals with European breeds in beef production systems and dual purpose systems as well [18]. A major contribution of Zebu breeds is to the composite breeds, such as Santa Gertrudis, Brangus, Beefmaster, Simbrah, Braford, and Charbray [1]. In Mexico, Brahman, Red Brahman, Nellore, Guzerat, Indubrazil, Gyr and Sardo Negro, are the most important representative Zebu breeds.

Results from the present study indicated low or null segregation of CAPN316 favorable genotypes in all studied Zebu breeds (Table 1); conversely, a more equilibrated occurrences of favorable C, and T alleles, were present for CAPN4751 and CAST-T1 loci, respectively (Table 1). The HWE test showed significant deviations in CAPN316 genotypes in all breeds ($P \le 0.005$). In contrast, Gyr was the only breed showing deviations from the HWE in the CAPN4751 locus. CAST-T1, exhibited no deviations from the HWE.

Some reports support the relative null and low frequencies of favorable allele segregating in Zebu, specifically in Brahman and Nellore breeds [9,10,15] (Table 2). There is only one study reporting CAPN316 and CAPN4751 frequencies in a Brahman cattle population in Mexico. Parra-Bracamonte et al. [15] reported relatively high

frequencies of favorable C alleles in both loci with a high occurrence in heterozygous genotypes. However, the results of the present study clearly indicate an almost null frequency of allele C in CAPN316 locus and a lower occurrence of favorable allele in CAPN4751 compared to that previous assessment clarifying the actual situation of Brahman populations from Mexico. For the other four breeds the authors did not find previous reports.

Theoretically, using validated SNPs for selection may represent the state of the art of genetic improvement for a trait that is very difficult or expensive to measure [8,19]. There is strong validating evidence of the usefulness of CAPN and CAST-T1 for the improvement of meat tenderness [7,8,9,10,16]. Additionally, Tait et al. [12,13] in recent research, evaluated the CAST-T1 effects on slice shear force in crossbred [12] and angus purebred cattle [13], and reported a significant improvement of shear force residual variance associated to the selection the favorable genotype, consequently preventing the risk of tough meat and strongly supporting the use of CAST-T1 polymorphism for marker assisted management or marketing of beef products.

Frequencies and distribution of favorable alleles for meat tenderness in the Zebu breeds considered in this study, at least for CAPN4751 and CAST-T1, suggest the possibility of using mating strategies to increase the frequency of the favorable alleles. For instance, in the Brahman breed, the most numerous sampled population, CAPN4751 and CAST-T1 haplotype combination showed an availability of 54% of animals with two favorable alleles and 6% of animals with three favorable alleles for meat tenderness. This information suggests the possibility to manage these available segregation frequencies to positively change meat toughness assisting current breeding objectives used in Brahman breed by selection of favorable allele carriers.

Despite the fact that the beef market in Mexico does not consider meat tenderness as an indicator for meat quality with no incentive for genetic improvement of this trait, since beef from Zebu cattle is considered tough, the implementation of strategies such as the one reported here may contribute to improve the perception of Zebu beef in some market niches and change the consumer perception. However, the application of measures such as Warner–Bratzler shear force in sire evaluation, as included for instance by the American Brahman Breeders Association in their breeding objectives [20], is comparatively more difficult in Mexico due to the absence of suitable infrastructure and other particularities of the beef cattle breeding sector. Therefore, the use of validated genetic markers may represent a more comprehensive strategy for the genetic improvement of meat tenderness in beef cattle, at least in the short term.

Economic returns for the implementation of introgression of the favorable alleles of the CAPN and CAST-T1 SNP markers have been

Table 2Literature reported genotype and allele frequencies of Calpain and Calpastatin polymorphisms in Zebu breeds.

Locus	Favorable allele	Breed	Genotype %			n	Frequency		Ref
			0	1	2		Favorable allele	Unfavorable allele	
CAPN316 C	С	Brahman	97	3	0	470	0.02	0.98	[7]
		Brahman	96	4	0	674	0.02	0.98	[8]
		Brahman	94	6	0	382	0.03	0.97	[10]
		Nellore	98	2	0	638	0.08	0.92	[11]
		Brahman	0	92	8	85	0.46	0.54	[15]
CAPN4751 C	С	Brahman	81	19	0	471	0.10	0.90	[7]
		Brahman	88	11	1	674	0.06	0.94	[8]
		Nellore	0	21	79	114	0.105	0.895	[9]
		Brahman	0	10	90	382	0.05	0.95	[10]
		Nellore	68	29	3	638	0.18	0.82	[11]
		Brahman	0	99	1	85	0.49	0.51	[15]
CAST-T1	T	Brahman	18	50	32	768	0.57	0.43	[8]
		Brahman	11	46	43	674	0.66	0.34	[8]
		Nellore	23	43	34	114	0.557	0.443	[9]
		Brahman	11	47	42	382	0.66	0.34	[10]
		Brahman	6	44	50	444	0.72	0.28	[16]

Ref: Reference. n: Sample size.

estimated by Weaber and Lusk [21]. Implementing a simulation assessment on the use of genetic markers for meat tenderness, Weaber and Lusk [21] indicated that a strategy in which bulls from upper 30% of genetic merit are selected each year would result in increased profitability of \$9.60/head for feeder cattle and \$1.23/head for fed cattle for 20 years. This significant economic benefit and the expected reduction of genotyping costs open the possibility of considering marker assisted selection or management as an actual option to improve meat tenderness.

In conclusion, the frequency of favorable alleles in Calpain genetic markers is low in the five Zebu breeds assessed in this study. Although the indirect improvement of meat tenderness by selection of tenderness segregating favorable alleles in Calpain loci would be limited, favorable alleles could be introgressed in target populations, making of CAPN4751 and CAST-T1 selection, a feasible alternative. Genotyping availability and cost reduction make this strategy a possibility to implement a marker assisted selection program and management in registered Zebu cattle.

Conflict of interest

The authors declare no conflicts of interest.

Acknowledgments

First author wants to thank Dr. Xianwu Guo and Prof Nicolas Lopez-Villalobos, for their comments and suggestions on the manuscript.

References

- Rischkowsky B, Pilling D. The state of the world's animal genetic resources for food and agriculture. Rome: Food and Agriculture Organization of the United Nations; 2007[http://www.fao.org/docrep/010/a1250e/a1250e00.htm].
- [2] Hansen PJ. Physiological and cellular adaptations of Zebu cattle to thermal stress. Anim Reprod Sci 2004;82-83;349-60. http://dx.doi.org/10.1016/j.anireprosci.2004.04.011.
- [3] Koohmaraie M. Biochemical factors regulating the toughening and tenderization process of meat. Meat Sci 1996;43:193–201. http://dx.doi.org/10.1016/0309-1740(96)00065-4.
- [4] Rubensam JM, Felício PE, Termignoni C. Influência do genotipo Bos indicus na actividade de calpastatina e na textura da carne de novilhos abatidos no sul do Brasil. Food Sci Technol 1998;18:2–6. http://dx.doi.org/10.1590/S0101-20611998000400009.
- [5] Delgado EJ, Rubio MS, Iturbe FA, Méndez RD, Cassís L, Rosiles R. Composition and quality of Mexican and imported retail beef in Mexico. Meat Sci 2005;69:465–71. http://dx.doi.org/10.1016/j.meatsci.2004.10.003.

- [6] Page BT, Casas E, Quaas RL, Thallman RM, Wheeler TL, Shackelford SD, et al. Association of markers in the bovine CAPN1 gene with meat tenderness in large crossbred populations that sample influential industry sires. J Anim Sci 2004;82:3474–81.
- [7] White SN, Casas E, Wheeler TL, Shackelford SD, Koohmaraie M, Riley DG, et al. A new single nucleotide polymorphism in CAPN1 extends the current tenderness marker test to include cattle of *Bos indicus*, *Bos taurus*, and crossbred descent. J Anim Sci 2005;83:2001–8.
- [8] Van Eenennaam AL, Li J, Thallman RM, Quaas RL, Dikeman ME, Gill CA, et al. Validation of commercial DNA tests for quantitative beef quality traits. J Anim Sci 2007;85: 891–900. http://dx.doi.org/10.2527/jas.2006-512.
- [9] Curi RA, Chardulo LA, Mason MC, Arrigoni MD, Silveira AC, De Oliveira HN. Effect of single nucleotide polymorphisms of *CAPN1* and *CAST* genes on meat traits in Nellore beef cattle (*Bos indicus*) and in their crosses with *Bos taurus*. Anim Genet 2009;40: 456–62. http://dx.doi.org/10.1111/j.1365-2052.2009.01859.x.
- [10] Smith T, Thomas MG, Bidner TD, Paschal JC, Franke DE. Single nucleotide polymorphisms in Brahman steers and their association with carcass and tenderness traits. Genet Mol Res 2009;8:39–46. http://dx.doi.org/10.4238/vol8-1gmr537.
- [11] Pinto LFB, Ferraz JBS, Meirelles FV, Eler JP, Rezende FM, Carvalho ME, et al. Association of SNPs on CAPN 1 and CAST genes with tenderness in Nellore cattle. Genet Mol Res 2010;9:1431–42. http://dx.doi.org/10.4238/vol9-3gmr881.
- [12] Tait RG, Shackelford SD, Wheeler TI, King DA, Keele JW, Casas E, et al. CAPN1, CAST, and DGAT1 genetic effects on preweaning performance, carcass quality traits, and residual variance of tenderness in a beef cattle population selected for haplotype and allele equalization. J Anim Sci 2014;92:5382–93. http://dx.doi.org/10.2527/jas.2014-8211.
- [13] Tait RG, Shackelford SD, Wheeler TL, King DA, Casas E, Thallman RM, et al. μ-Calpain, Calpastatin, and growth hormone receptor genetic effects on preweaning performance, carcass quality traits, and residual variance of tenderness in Angus cattle selected to increase minor haplotype and allele frequencies. J Anim Sci 2014;92:456–66. http://dx.doi.org/10.2527/jas.2013-7075.
- [14] Allan MF, Smith TPL. Present and future applications of DNA technologies to improve beef production. Meat Sci 2008;80:79–85. http://dx.doi.org/10.1016/j.meatsci.2008.05.023.
- [15] Parra-Bracamonte GM, Sifuentes-Rincón AM, Cienfuegos-Rivas ME, Tewolde-Medhin A, Martínez-González JC. Polimorfismo en el gen de la μ-calpaína en ganado Brahman de registro de México. Arch Latinoam Prod Anim 2007;15:33–8.
- [16] Casas E, White SN, Wheeler TL, Shackelford SD, Koohmaraie M, Riley DG, et al. Effects of Calpastatin and μ-calpain markers in beef cattle on tenderness traits. J Anim Sci 2006:84:520–5.
- [17] Rousset F. Genepop'007: A complete reimplementation of the Genepop software for Windows and Linux. Mol Ecol Resour 2008;8:103–6. http://dx.doi.org/10.1111/j.1471-8286.2007.01931.x.
- [18] McDowell RE, Wilk JC, Talbott CW. Economic viability of crosses of Bos taurus and Bos indicus for dairying in warm climates. J Dairy Sci 1996;79:1292–303. http://dx.doi.org/10.3168/jds.S0022-0302(96)76484-6.
- [19] Dekkers JCM, Hospital F. Multifactorial genetics: The use of molecular genetics in the improvement of agricultural populations. Nat Rev Genet 2002;3:22–32. http://dx.doi.org/10.1038/nrg701.
- [20] Garrick DJ. The nature, scope and impact of genomic prediction in beef cattle in the United States. Genet Sel Evol 2011;43:1–11. http://dx.doi.org/10.1186/1297-9686-43-17.
- [21] Weaber RL, Lusk JL. The economic value of improvements in beef tenderness by genetic marker selection. Am J Agric Econ 2010;92:1456–71. http://dx.doi.org/10.1093/ajae/aaq062.