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ADVANCES IN THE IDENTIFICATION OF CANDIDATE GENES FOR SELECTION OF CATTLE FOR RIBEYE AREA: A SYSTEMATIC REVIEW

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ABSTRACT - In this systematic review, we aimed to clarify which genes or genomic regions influence the development of the *longissimus dorsi* muscle in cattle. An integrative review was performed based on quantitative and qualitative data published on PubMed, using the search criteria "ribeye area and carcass and taurus". We verified significant influence of the breed origin, so that, provided environmental effects that benefit the formation of muscle tissues, with the proportional allometric growth, the fat deposition will not be affected; however, there are nutritional limitations for the development of all tissues in commercial livestock production systems. The diversity of genes and SNPs associated with loin eye area (REA) allow the conduction of investigations within breed, associating REA with traits of economic importance that are difficult to measure, such as those related to reproduction. The processes associated with the expression of the genes reported in this review represent a frontier that will be crossed by the omics sciences.
Keywords: BTA 14, Hanwoo, *longissimus dorsi*, yield index.

AVANÇOS NA IDENTIFICAÇÃO DE GENES CANDIDATOS À SELEÇÃO DE BOVINOS PARA ÁREA DE OLHO DE LOMBO: UMA REVISÃO SISTEMÁTICA

RESUMO - Nesta revisão sistemática, objetivamos esclarecer quais genes ou regiões genômicas influenciam o desenvolvimento do conjunto de músculos denominado *longissimus dorsi*, em bovinos. Uma revisão integrativa foi realizada com base em dados publicizados de forma quantitativa e qualitativa com todas as publicações da PubMed, utilizando os critérios de busca "ribeye area and carcass and taurus". Constatou-se que há significativas influências de origem racial, de modo que, sendo proporcionados efeitos ambientais que favoreçam a formação dos tecidos musculares, com o devido crescimento alométrico, não haverá prejuízo na deposição de gordura; todavia, é presumível haver limitações nutricionais para o desenvolvimento de todos os tecidos em sistemas comerciais de exploração. A diversidade de genes e SNPs associados projetam lastro para realização de investigações, individualizados por raça, associando AOL com características de importância econômica difíceis de mensurar, como aquelas relacionadas à reprodução. Os processos associados à expressão dos genes citados nesta revisão projetam-se como fronteira a ser percorrida pelas ciências ômicas.
Palavras-chaves: BTA 14, Hanwoo, *longissimus dorsi*, índice de rendimento.

INTRODUCTION

Human feeding has been always associated with the development of production technologies, processing, and consumption. In this sense, beef was obtained through extractivism for thousands of years, until it became a product of several processes of breeding, slaughter, and commercialization. The consumption was intensified by the population growth and made most breeders adjust their livestock production systems.

When animals, such as cattle, are slaughtered, the carcasses are evaluated in the cold storage environment, where the variation in the proportion of edible parts varies significantly between categories, and even between individuals of the same breed and category. These particularities are indicators of the profitability of cattle production, as well as the efficiency of genotypes in expressing nutritional phenotypes. Do et al. (2016) indicate that qualitative evaluation is associated with consumer perception, with some animals being attributed a superior ability to constitute better carcasses.

For decades, the perimeter of the diameter of the *longissimus dorsi* muscle has been measured during the slaughter, through a cut between the 12th and 13th ribs of the left side of the carcass. The outline of this measurement is commonly used to estimate the REA (ribeye area) or LEA loin eye area (FELICIO, 2018). REA data obtained *in vivo* have been used in the routine of several beef cattle farms, in order to help in the genetic selection and culling processes.

Chung et al. (2020) clarify that carcass characteristics in cattle are constructed under the influence of genetic and non-genetic factors, in which the heritability of REA characteristics, carcass weight and marbling are more accurately estimated when considering regional particularities, such as temperature and humidity; with a predicted genotype-environment interaction for these characteristics.

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The genetic basis of REA has been studied for decades and variations have been found between breeds and between animals of the same breed for this trait. This variation justifies the selection of breeding animals based on data of the phenotype REA, along with information of pedigree and/or molecular markers. Single nucleotide polymorphisms (SNP) associated with carcass characteristics allow genetic selection for improvements in REA, however, the percentage of variation explainable by known polymorphisms is variable even within a single genetic group, with some breeds having a limited number of progenitors in greater evidence, architecting the formation of current herds, with records of herds of hundreds of thousands of animals and significant relationships being identified (MUDADU et al., 2016).

The phenotypic expression of REA has a complex influence of several environmental and genetic variables. Furthermore, there are genetic and physiological interactions between REA and other phenotypes, which are strongly influenced by the production systems.

In this context, a systematic literature review was carried out to clarify which genes or genomic regions influence the development of the *longissimus dorsi* muscle in cattle.

METHODOLOGY

Systematic review

An exploratory search was conducted using the Integrative Review methodology based on published data, quantitative or qualitatively, according to Pollock and Berge (2018). The definition of the term numbers is associated with the minimum number of keywords constructed in academic works, where three to five terms tend to present a minimum range of capture of publications on the subject, delimiting the subject. It is considered that the definition of the terms boosts the identification of articles with potential interest, with subsequent steps being foreseen that include the effective analysis of the results. The indexing base number does not necessarily lead to gains in identifying journals. The success of the systematic review is associated with the definition of the problem; there must be not only a volume of publications but also social and economic relevance of the research object, as well as being an evolving subject of scientific discoveries.

Given the problematic, the next step consisted of defining the criteria of inclusion or exclusion. The English language was selected due to the great number of scientific publications in this language. Subsequently, we chose the database for indexation. Considering that, the effectiveness of the process of synthesis is in the technical compilation of the main publications and not in the huge volume of articles, it was essential that the database included the main journals. In this case, some of the most prominent journals in the field of Animal Sciences are Journal of Animal Science, PlosOne, Animals, Frontiers in Genetics, and BMC Genomics.

A strategy of using more than one database is acceptable; however, sometimes this strategy does not result in gains of efficiency for obtaining manuscripts. Other systematic reviews considered the database selected in the current study (PubMed) along with others. This resulted in a significant number of duplicate papers so that 42% of the publications obtained using more than one database had to be removed (BAIAK et al., 2019). In this context, using a single and appropriate database for indexation seems the most efficient strategy. This is corroborated by the fact that the PubMed database has only two main knowledge fields, which include important journals in the field of Agrarian Sciences (UFU, 2016).

The PubMed (PMC) database was selected accessing the Federated Academic Community (CAFe) that is maintained by the Brazilian National Research and Educational Network (RNP), via the Federal University of Piauí (UFPI). The PMC database is organized by the National Center for Biotechnology Information, 8600 Rockville Pike, Bethesda MD, 20894, EUA. The database related to the field of Health is the search engine for accessing the MEDLINE (Medical Literature Analysis and Retrieval System Online), which indexes more than 5,000 journals with content related to biotechnologies.

Literature search strategy

The advanced module of the search system was selected and the following terms were used: “rib eye area” (name of the variable); “carcass” (the group of traits of interest); and “taurus” (species of interest) to obtain the articles actually considered (FIGURE 1).

The terms were submitted to verification in all fields, in order to show the papers that could be selected. All references were exported to the Mendeley Desktop software v1.19.4, adjusted for the Brazilian norms (NBR) 10520.

The relevance of the selected papers was based on the use of the diameter of the *longissimus dorsi* muscle, provided that REA was the main or secondary trait used in the study. In this step, articles that had not a minimum content about REA in cattle were discarded.

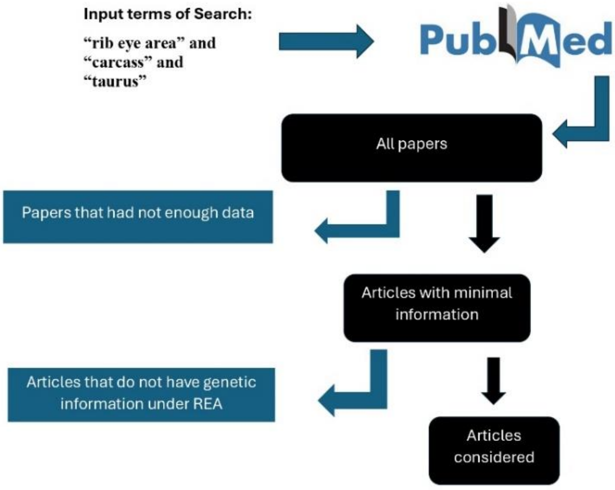

FIGURE 1 - Dynamics of search and selection of manuscripts. Source: authors.

RESULTS AND DISCUSSION

The search resulted in 103 articles, which were read in fact. Subsequently, papers that had not enough data, had not a minimum of information about the genetic basis of REA in cattle or did not show evidence that contributed to clarifying the genetic origin of this trait were removed. Therefore, 48 articles were used for the following discussion.

There are few variations of REA among animals of breeds with similar frames: for example, Angus, Wagyu, and Jersey. No significant differences were found among these breeds; however, Angus cattle showed the lowest subcutaneous fat and the fattiest kidney ($p = 0.01$) (JABOREK et al., 2019).

Based on more than 150,000 records of REA collected by certified technicians, the variations among animals were very expressive. At least 13.8 standard deviations were observed in all breeds. The mean REA of animals of continental breeds such as Simmental (86 cm^2) was higher than that observed in British breeds, such as Angus (78.2 cm^2) (SCHMIDT et al., 2020). Crossings are used for the manipulation of REA in cattle. Animals of the Brangus breed, for instance, have mean REA significantly higher (81.14 cm^2) than zebu cattle that were not crossed with taurines (73.23 cm^2), which showed the lowest REA ($p\text{-value} \leq 0.05$) (FLOWERS et al., 2018).

The REA trait has the lowest number of genes involved in its expression (from 167 to (LEE et al., 2020), in comparison to phenotypes such as fat thickness, carcass weight, and marbling (JABOREK et al., 2019).

The influence of genes on REA is more evident during the slaughter age of the animals; however, since the embryonic phase, it is possible to identify differences among individuals as a function of their genetic differences. In this case, parental genomes explained the higher proportion of the variation in the size of myofibrils of the *longissimus dorsi* muscle. The paternal genome explained the higher variation of relative weight and the lower variation of the absolute weight ($p\text{-value} \leq 0.05$). More than 20% of the variation was due to the gender of the offspring (XIANG et al., 2013).

The Fas gene (APO-1, TNFRSF6), located in the bovine chromosome 26 (BTA26), is associated with beef quality. Hanwoo cattle that have the Faz gene have higher deposition of intramuscular fat (KIM et al., 2016).

In genome-wide association studies, the bodyweight is the continuous variable most clearly associated with genes or sets of genes with higher relevancy to explain the genetic and phenotypic variances of the animals. In this sense, single nucleotide polymorphisms (SNPs) BTB-01280026 and Hapmap27934-BTC-065223, located on the chromosome BTA14, are the main polymorphisms within a group of dozens of SNPs. Nevertheless, there were no SNPs strongly associated with REA. According to Lee et al. (2017), later studies also showed associations in the chromosome BTA14 and included the BTA5 too. Neither BTA14 nor BTA5 is among the chromosomes more frequently studied for REA in cattle (LEE et al., 2020).

Groups of genes present on chromosomes BTA 1, 6, 9, and 19 contributed to explaining, respectively, 2.27, 1.00, 2.22, and 1.00% of the genetic variation (σg) of REA. The chromosome BTA 14 showed the group of genes that gave the highest contribution (7.98% of σg), namely: XKR4, TMEM68, TGS1, LYN, RPS20, MOS, PLAG1, CHCHD7, SDR16C5, SDR16C6, PENK, and LOC101907667 (NASERKHEIL et al., 2020).

The genes XKR4, TMEM68, LYN, PLAG1, CHCHD7, and PENK are also associated with carcass weight and weight at 1 year of age in cattle. Some of these genes are also associated with fat thickness. The expression of the TMEM68 gene is determined by microRNAs (miRNAs). The genes LYN and PENK influence physiologically the protein linkage. In addition, LYN regulates metabolic processes of lipids and interactions with the PECAM1 gene, which influence the feed intake, growth, and meat tenderness in cattle. The PLAG1 gene is involved in the growth of all multicellular organisms, influence the stature and meat tenderness in some cattle breeds (NASERKHEIL et al., 2020).

Mudadu et al. (2016) conducted a survey before research using Genome-Wide Association Studies (GWAS), to find genes associated with REA in the literature. Genes such as TBC1D5, PPFIA2, PCDH9, OTP, OPN5, MAF, LOC786312, LOC618866, LOC528040, LOC516179, LOC515830, and GPR12 were indicated as probable influencers of REA. However, those genes showed a low significance rate. Among the genes mentioned above, all those associated with REA were also related to other traits.

Some analyses can also be performed using measurements derived from REA. Polymorphisms of the DNMT1 gene affected significantly the width (DNMT3a) and length (DNMT3b) of the rib-eye (LIU et al., 2015). The markers ARS-BFGL-NGS-39006 and ARS-BFGL-NGS-100046 represented, respectively, 2.6 and 3.01 of the genotypic variation; whereas the markers NCBI rs109741381 and rs110527834 explained, respectively, 1.35 and 0.70 of the phenotypic variation of REA (SUDRAJAD et al., 2016).

Nevertheless, some physiological processes were more clearly observed. The response to peptide, response to insulin, response to organonitrogen compound, response to peptide hormone, and positive regulation of small molecule metabolic process represented, respectively, 14.1, 11.3, 19.7, 12.7, and 9.9% of the GeneRatio (LEE et al., 2020).

The diversity of candidate genes hinders studies that aim to rank these genes based on their effects. Another systematic review identified the frequency in which some genes were cited in the scientific literature, in order to identify those genes that had been reported at least 30 times. For most of the carcass traits, the minimum number of genes was identified, except for REA. Therefore, the studies that investigated the genes that influence REA were not enough to go over the pleiotropy and synergisms of genes necessary for the construction of this phenotype (LEE et al., 2020).

Thus, pleiotropic genes influence the REA trait, so that the genetic variation of this trait is highly spread throughout the genetic material, which results in some variation among breeds. Animals of the Charolais breed, for example, have a lower number of quantitative trait loci (QTL) involved in the expression of REA, in comparison to Hereford cattle. On the other hand, the chromosome BTA2 has a higher influence on REA in Limousin and Red Angus cattle (SAATCHI et al., 2014).

Heritability

The procedures involved in the collection of phenotypes in animals have different levels of difficulty. In the case of REA, the measurement can be performed without the use of expensive resources, with laboratory analysis that can be carried out using samples available electronically (HERAS-SALDANA et al., 2019).

The loin eye area is one of the bovine traits that have the higher heritability (h^2), with influent SNPs spread throughout all chromosomes (MUDADU et al., 2016). Fernandes Júnior et al. (2016) demonstrated that the genetic component represents a considerable but lower proportion of the phenotypic variation of REA. The part of the variance that can be transmitted along the generations (additive genetic variance) is 10.88, which indicates that there are heritability estimates that range from moderate to high magnitude for REA, as shown in Table 1.

TABLE 1 - Heritability estimates (h^2) for loin eye area (REA) in cattle.

Breed	N	h^2	Mean REA (cm^2)	Authors
Hanwoo	72,969	0.23	81.8	Do et al. (2016)
Taurine	1366	0.35	86.74	Ekine-Dzivenu et al. (2017)
Hanwoo	5,218	0.43	82.99	Mehrban et al. (2017)
Hanwoo	1011	0.41	81.1	Lee et al. (2017)
Nellore	2283	0.44	51.4	Ceacero et al. (2016)
Hanwoo	10,215	0.33	95.8	Chung et al. (2020)
Angus	281,982	0.32	78.2	Schmidt et al. (2020)
Hereford	49,602	0.35	70.6	Schmidt et al. (2020)
Simmental	59,576	0.41	86	Schmidt et al. (2020)
Hanwoo	1262	0.42	77.85	Lee et al. (2017)
Red Angus	39,227	0.41	71.06	Boldt et al. (2018)
Nellore	669	0.55	60.45	Mudadu et al. (2016)

N: sample size.

The heritability estimates for REA, which is rarely lower than 0.30, can be influenced by the breed (SCHMIDT et al., 2020). REA has one of the highest heritability estimates, with associated SNPs spread throughout the genome (MUDADU et al., 2016). Rib eye area (REA) has high heritability (h^2=0.66), and selection for this could yield genetic gains for other traits with lower heritability, such as carcass weight (WANG et al., 2023).

The phenotypic variation of REA is more explained genes with additive effect (0.46 ± 0.08) in purebred animals (Angus, Charolais, Hereford, Simmental, and Limousin) than in crossbred individuals (0.38 ± 0.03). This is also valid for the effects of dominance, which is better explained in purebred cattle (AKANNO et al., 2018).

Selection

In Hanwoo cattle with a selection differential of 3.34 was possible to obtain an average annual increase of 0.27 cm^2 of REA as a genetic response to selection (PARK et al., 2013), as shown in Table 2. Furthermore, in the herd reported in the study mentioned above the carcass weight gain was simultaneously maintained for approximately 15 years.

TABLE 2 - Evolution of carcass weight, loin eye area (REA), and marbling in a herd of Hanwoo cattle.

Trait	Years							
	2001	2002	2004	2005	2006	2008	2009	2010
Carcass weight (kg)	345	358	379	378	385	405	413	419
REA (cm2)	79.6	81.1	83.1	81.5	83.1	86.5	88.0	88.7
Marbling	3.4	3.5	4.2	4.9	4.8	5.0	5.3	5.2

Adapted from Park et al. (2013).

Since 1974 there is an evolution in the bodyweight of the animals of the breeding program of Hanwoo cattle. In addition, since 2010 they have also observed the evolution of REA. In the case of the Korean Hanwoo cattle, the breeding has been directed towards the control of backfat, which was recently on average 8.6 mm (PARK et al., 2013), that is considered an excessive value for this phenotype. Some authors observed that adult males not castrated (n = 88,000) of different breeds reared under Australian tropical conditions tend to have larger REA (83 cm^2), whereas females of the same age reared under the same conditions (n = 101,000) have lower REA (62.9 cm^2) (BOERNER et al., 2014).

When different mixed models (GBLUP, BayesL, and six types of BayesC) were used to predict the genetic merit of genotyped Hanwoo beef cattle for REA, the model GBLUP had the highest accuracy when the sample sizes represented half of the original training set. The BayesL model outperformed GBLUP when three-fourths of the original training set was used. When the whole population was used, the model BayesC97 outperformed GBLUP. Therefore, the models mentioned above have different accuracies regarding the backfat thickness, carcass weight, REA, and marbling score (MEHRBAN et al., 2017).

CONCLUSIONS

Correlations of REA with body mass are positive linear. The influence of REA on fat thickness relies on the breed origin. Apparently, if the environmental effects are favorable to the formation of muscle tissues, with the appropriate allometric growth, the fat deposition will not be affected. Nevertheless, there are nutritional limitations for the development of all tissues in commercial livestock production systems.

Part of the feed efficiency can be better understood based on measurements obtained using ultrasonography. In this sense, the diameter of the *longissimus dorsi* muscle can be partially used as an indicator of the body composition.

The diversity of genes and SNPs associated with REA allow the conduction of studies within each cattle breed, associating REA with other traits of economic importance that are more difficult to measure, such as those related to reproduction. The processes related to the expression of the genes mentioned in the current review represent a frontier that will be crossed by the omics sciences.

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