GABRIELA CORDEIRO BRILHANTE

GENETIC PARAMETERS FOR FERTILITY INDICATOR TRAITS, BIOMETRIC MEASUREMENTS AND REPRODUCTIVE INDICES IN TABAPUÃ CATTLE

LAVRAS – MG
2023

GABRIELA CORDEIRO BRILHANTE

GENETIC PARAMETERS FOR FERTILITY INDICATOR TRAITS, BIOMETRIC MEASUREMENTS AND REPRODUCTIVE INDICES IN TABAPUÃ CATTLE

Dissertação apresentada à Universidade Federal de Lavras como parte das exigências do Programa de Pós Graduação em Zootecnia para obtenção do título de Mestre

Orientadora: Profª. Dra. Sarah Laguna Conceição Meirelles
Co-orientador: Prof. Dr. José Camisão de Souza

LAVRAS – MG
2023

GABRIELA CORDEIRO BRILHANTE

GENETIC PARAMETERS FOR FERTILITY INDICATOR TRAITS, BIOMETRIC MEASUREMENTS AND REPRODUCTIVE INDICES IN TABAPUÃ CATTLE

PARÂMETROS GENÉTICOS PARA CARACTERÍSTICAS INDICADORAS DE FERTILIDADE, MENSURAÇÕES BIOMÉTRICAS E ÍNDICES REPRODUTIVOS EM FÊMEAS TABAPUÃ

> Dissertação apresentada à Universidade Federal de Lavras como parte das exigências do Programa de Pós Graduação em Zootecnia para obtenção do título de Mestre

APROVADA EM 04 DE ABRIL DE 2023.

Dra. Sarah Laguna Conceição Meirelles UFLA

Dra. Alessandra Alves Silva Unesp/FCAV

Dr. Rilke Tadeu Fonseca de Freitas UFLA

Orientadora
Dra. Sarah Laguna Conceição Meirelles

LAVRAS – MG 2023

AGRADECIMENTOS

RESUMO GERAL

A busca por identificação de animais precoces vem avançando ao longo dos anos, devido ao intenso crescimento populacional e aumento da demanda por proteína de origem animal. A exigência do mercado de animais melhoradores é imprescindível para os avanços pecuários, porém algumas características não são simples de selecionar. A resposta à seleção de características reprodutivas é mais lenta, pois geralmente elas possuem baixa herdabilidade, sendo muito influenciadas por fatores ambientais. Com o intuito de acelerar o melhoramento genético e a seleção dessas características reprodutivas, a busca por características indicadoras de fertilidade de forma precoce e de fácil mensuração é interessante, facilitando assim o processo de seleção a campo. O objetivo deste trabalho foi buscar características indicadoras de fertilidade em fêmeas zebuínas, estimar as herdabilidades e avaliar as correlações genéticas entre as mesmas. Foram coletados dados de 934 fêmeas, nascidas entre 1993 a 2020, nos quais foram avaliadas diversas características como tamanho vulvar, contagem de folículos antrais (CFA), concentração do hormônio Anti-Mulleriano (AMH), medidas biométricas e índices reprodutivos do Programa de melhoramento genético de zebuínos (PMGZ) da raça Tabapuã. Os parâmetros genéticos das características foram estimados via inferência bayesiana utilizando executáveis da família BLUPF90. As análises foram realizadas utilizando modelo misto linear que incluiu os efeitos de grupo de contemporâneo formado por ano de nascimento, fazenda e paridade, além dos efeitos de classe de idade e classe de peso. O efeito aleatório do próprio animal também foi considerado. Os valores de herdabilidade obtidos para habilidade mais provável de produção (0,88), altura do anterior (0,42), perímetro torácico (0,37), comprimento da rima (0,32), idade ao primeiro parto (0,28), contagem folicular antral (0,24) e largura da vulva (0,20) demonstram que são características responsivas à seleção direta. De acordo com as estimativas de correlações genéticas, conclui-se que não há correlação genética entre as características estudadas.

Palavras-chaves: comprimento da rima; correlação genética; herdabilidade; largura vulvar.

GENERAL ABSTRACT

Over the years, the search for identification of precocious animals has been advancing due to the intense population growth and increased demand for animal protein. The demand from the market for breeding animals is essential for livestock advances, but some traits are not easy to select. The response to the selection of reproductive traits is slower, as they generally have low heritability, being greatly influenced by environmental factors. With the aim of accelerating the genetic improvement and selection of these reproductive traits, the search for early and easily measured fertility indicator traits is interesting, facilitating the field selection process. The aim of this work was to look for traits that indicate fertility in Zebu females, estimate the heritability and evaluate the genetic correlation between them. We collected data from 934 females, born between 1993 and 2020, evaluating traits such as vulvar size, antral follicle counts (AFC), concentration of Anti-Mullerian hormone (AMH), biometric measurements and reproductive indices of the breeding program of Zebu cattle (PMGZ) of the Tabapuã breed. The genetic parameters of the traits were calculated by Bayesian analysis using executables from the BLUPF90 family. The analyzes were carried out using a linear mixed model that included the effects of contemporary group formed by year of birth, farm and parity, in addition to the effects of age and weight class. The random effect of the animal itself was also considered. The heritability values obtained for most probable producing ability (MPPA) (0.88), anterior height (AH) (0.42), thoracic depth (TD) (0.37), rima length (RIMA) (0.32), age at first calving (AAFC) (0.28), antral follicle count AFC (0.24) and vulva width (VW) (0.20) demonstrate that they are traits responsive to direct selection. According to the estimates of genetic correlations, it can be concluded that there is no genetic correlation between the traits studied.

 Keywords: genetic correlation; heritability; rima length; vulvar width.

RESUMO GRÁFICO

Resumo gráfico. Os animais foram avaliados quanto ao tamanho da genitália externa, contagem folicular antral (CFA), medidas biométricas, concentração de hormônio Anti-Mulleriano (AMH) e índices reprodutivos fornecidos pelo Programa de melhoramento genético de zebuínos (PMGZ). Foram estimadas as herdabilidades e as correlações genéticas entre essas características. A maioria delas respondem à seleção direta.

SUMÁRIO

PRIMEIRA PARTE – REFERENCIAL TEÓRICO

1 INTRODUÇÃO

O Brasil possui um dos maiores rebanhos bovinos do mundo com mais de 224 milhões de cabeças (IBGE, 2021), sendo atualmente um dos principais países produtores e exportadores de carne bovina no mundo (ABIEC, 2022; USDA, 2020), e apontado a continuar sendo uma das principais potências da pecuária de corte mundial nos próximos anos (EMBRAPA GADO DE CORTE, 2020; RODRIGUES & MARTA-COSTA, 2021).

Devido ao crescimento populacional, a demanda por proteína de origem animal vem aumentando e, consequentemente, os produtores precisam se tornar mais eficientes na produção de carne e leite, tendo a necessidade de selecionar indivíduos geneticamente superiores. Entretanto, a intensa seleção para características relacionadas à produção pode ter afetado negativamente a eficiência reprodutiva das fêmeas bovinas (MACULAN et al., 2018).

A identificação de fêmeas superiores quanto às características reprodutivas pode ser realizada pelas avaliações genéticas das matrizes, planejamentos de acasalamentos direcionados e uso de biotecnologias reprodutivas, em sistema com elevada pressão de seleção. A seleção direta para fertilidade pode se tornar complexa, isso devido a fatores como os ambientais, aos longos períodos de avaliações de desempenho das fêmeas, como também a baixa herdabilidade dessas características (ZINK et al., 2011). Portanto, a identificação de características mais facilmente mensuráveis, e que estão correlacionadas com status de maior fertilidade nesses animais se faz necessária, podendo as matrizes serem avaliadas de maneira mais precoce no sistema de produção (SILVA et al., 2005).

Alguns trabalhos apontam que o tamanho de estruturas reprodutivas pode ser utilizado como indicadores de fertilidade, através das correlações fenotípicas estimadas (MESQUITA et al., 2016; MACULAN et al., 2018), além disso, já comprovaram que o tamanho dos ovários, é um bom preditor da reserva folicular e da funcionalidade ovariana (GRIGOLETTO et al., 2020; IRELAND et al., 2008; MACULAN el al., 2018; MODINA et al., 2014).

A estimativa do tamanho da reserva folicular é realizada de maneira indireta, tendo como base a contagem dos folículos antrais (CFA) nos ovários, entretanto tal técnica apresenta um custo oneroso, devido a necessidade de mão de obra qualificada e uso da

ultrassonografia. Com isso, encontrar características que consigam predizer essa CFA de uma forma mais simples, prática, barata e precoce é essencial para que o processo do melhoramento genético seja mais eficiente.

Outro parâmetro utilizado como indicador de fertilidade é o hormônio Anti-Mulleriano (AMH), estando este fortemente associado à CFA (ALWARD e BOHLEN 2019; IRLAND et al., 2010; GRIGOLETTO et al., 2020; MACULAN et al., 2018). O AMH funciona como um modulador do desenvolvimento folicular, evitando a atresia folicular precoce e a depleção da reserva folicular pré-antral (MONNIAUX et al., 2012). Uma grande vantagem do uso do AMH como marcador de fertilidade, é a pequena variabilidade ao longo dos vários estágios dos ciclos reprodutivos e estral, porém essa técnica apresenta um elevado custo de mensuração (MACULAN et al., 2018).

Estudos recentes demonstraram que a concentração do AMH e o tamanho de estruturas reprodutivas (principalmente da vulva), podem ser correlacionadas fenotipicamente com reserva ovariana de fêmeas bovinas (MESQUITA et al., 2016; MACULAN et al., 2018; VASCONCELOS et al., 2020). Porém, até o presente momento, ainda não foi amplamente estudado se essas características possuem uma correlação genética, e se são responsivas à seleção.

O entendimento das relações entre marcadores de fertilidade e as características fenotípicas externas, podem levar ao desenvolvimento de técnicas de fácil aplicação e baixo custo, passíveis a serem aplicadas no processo de seleção para maximizar eficiência reprodutiva (MACULAN et al., 2018). Além disso, encontrando correlações favoráveis, os produtores poderão otimizar o sistema de produção, selecionando as fêmeas mais precocemente, aumentando o ganho genético e reduzindo o intervalo de gerações, permitindo assim uma maior rapidez do retorno do capital investido.

Dessa forma, o objetivo deste trabalho foi estimar as herdabilidades e as correlações genéticas entre tamanho vulvar, contagem de folículo antral, concentração de AMH e dados reprodutivos do PMGZ buscando características indicadoras de fertilidade.

2 REFERENCIAL TEÓRICO

2.1 Características da raça Tabapuã

O Tabapuã é uma raça brasileira, oriunda dos cruzamentos entre a raça Mocho Nacional de ancestralidade ibérica com as raças zebuínas Nelore, Guzerá e Gir, que são de origem indiana. A raça se originou no município de Tabapuã, no estado de São Paulo (SP) na década de 1940, mas somente em 1981, o Tabapuã foi reconhecido como uma raça pela Associação Brasileira de Criadores de Zebu (ABCZ) e, essa passou a se tornar encarregada de realizar os registros genealógicos (ABCT, 2003).

Algumas das principais qualidades da raça Tabapuã são o bom ganho de peso, docilidade, caráter mocho e fêmeas com boa habilidade materna, garantindo assim um bom desenvolvimento dos bezerros.

2.2 Medições vulvares

A metodologia de avaliação morfométricas da genitália externa de fêmeas bovinas foi descrita por Mesquita et al. (2016), sendo o comprimento da rima o comprimento entre as comissuras dorsais e ventrais da fenda vertical da vulva do animal, já a largura da vulva pode ser determinada pela distância entre as bordas laterais da vulva, a partir do ponto médio da rima em um ângulo de 90 graus.

Segundo Maculan et al. (2018), alguns aspectos como o escore de condição corporal, estado fisiológico, fase do ciclo reprodutivo e genética das fêmeas, podem interferir nos resultados das medições vulvares, conforme observado nas fêmeas da raça Tabapuã, onde a largura da vulva foi estatisticamente maior nas fêmeas de alta CFA (8,81±0,12), em relação às de média (8,42±0,11) e baixa CFA (8,38±0,13).

Maculan et al. (2018) analisando dados de fêmeas Tabapuã observaram que o comprimento de rima foi significativamente menor em nulíparas em comparação com vacas primíparas e multíparas, não havendo também efeito da paridade sobre a largura da vulva. Já em trabalho no qual Maculan (2018) analisou fêmeas Brahman, Angus e Simental, a largura vulvar em fêmeas nulíparas e primíparas foi inferior às multíparas, sendo observada a necessidade de mais estudos a respeito.

Alguns estudos de associações fenotípicas demonstraram correlações positivas entre medições vulvares e CFA, como no estudo realizado por Pinto (2018), que observou uma correlação significativa entre tais características nas raças Holandesa (r = 0,26) e Girolando (r = 0,53), trazendo como hipótese que os animais com vulva maior seriam mais férteis. Essa observação vai de encontro ao relatado por Maculan (2018) em análise de fêmeas zebuínas e taurinas, onde foi observado maiores números de ovócitos viáveis nas fêmeas da classe de largura vulvar grande (18,86 ±1,76; n=38), quando comparada à classe pequena (10,15 ±1,49; n=53).

2.3 Contagem folicular antral

Os folículos antrais, são folículos em estágio de desenvolvimento caracterizado pela existência da cavidade antral, sendo possível sua visualização via avaliação ultrassonográfica (COUTINHO et al., 2007). A quantificação desses folículos é denominada como contagem folicular antral (CFA). Em avaliações seriadas no mesmo animal, a CFA apresenta grande consistência, porém, existe grande variabilidade no número de folículos entre animais (BURNS et al., 2005; CUSHMAN et al., 2009; IRELAND et al., 2007). Dessa forma, seria possível a classificação das fêmeas segundo a CFA em qualquer fase do ciclo estral (MACULAN, 2015). Diante disso, a CFA já vem sendo utilizada como um critério de seleção para a identificação de animais superiores geneticamente para as características reprodutivas, sendo o método mais adotado para estimação da reserva folicular ovariana (BANCSI et al., 2002).

Segundo Morotti et al. (2015), a grande variabilidade da CFA, é atribuída a diferenças entre os rebanhos, categorias, manejo, sanidade, alimentação e a falta de padronização nos critérios de seleção dos grupos, tornando-se complexo o conhecimento e a padronização da CFA sobre a fertilidade das fêmeas.

De acordo com alguns estudos (MACULAN, 2018; MACULAN et al., 2018; PINTO, 2018), a CFA apresenta correlações fenotípicas positivas com medidas morfométricas de genitália externa como a largura vulvar (0,16; 0,08; 0,26). Além disso, a CFA também está relacionada a uma maior recuperação de ovócitos e produção embrionária (IRELAND et al., 2007), sendo que tal correlação apresenta características vantajosas em relação a produção de embriões (MOROTTI et al., 2015)

Segundo Grigoletto et al. (2020), os níveis plasmáticos de AMH e a população de folículos antrais são características hereditárias em bovinos Nelore, podendo ser melhoradas

por meio de avaliação genômica e seleção genética, pois AMH e CFA são características altamente correlacionadas entre si geneticamente (rG= 0,81).

Mais estudos ainda são necessários para demonstrar como a CFA pode ser utilizada como uma característica indicadora de fertilidade, principalmente trabalhos voltados para a análise de parâmetros genéticos e o mérito genético desta característica.

2.4 Hormônio Anti-Mulleriano (AMH)

O AMH é uma glicoproteína dímera que desempenha importante função no sistema reprodutivo. Nas fêmeas, ele é produzido especificamente pelas células da granulosa dos folículos em desenvolvimento, controlando a velocidade do crescimento folicular por inibir a enzima aromatase (MORAIS et al., 2017; RICO et al., 2011). Este hormônio tem sido pesquisado em várias aplicações biotecnológicas, sendo que uma das mais relevantes já observadas é a relação positiva entre a sua concentração circulante e a CFA, sendo apontado como um indicador confiável desta característica (ALWARD et al., 2019; BATISTA et al., 2014; IRELAND et al., 2008; IRLAND et al., 2010 MACULAN, 2015; MONNIAUX et al., 2013; MORAIS et al., 2017; RICO et al., 2009).

Em fêmeas, o AMH atua como modulador do desenvolvimento folicular, prevenindo a atresia folicular precoce e a redução da reserva folicular pré-antral (MONNIAUX et al., 2012). Considerando o fato de que em mamíferos, as fêmeas já nascem com uma reserva de gametas pré-determinada (WALLACE e KELSEY, 2010), a avaliação da reserva folicular ovariana mede o potencial reprodutivo do indivíduo, isso em função da quantidade dos oócitos ainda viáveis em diferentes momentos da vida (MORAIS et al., 2017). Uma grande vantagem do uso do AMH como marcador de fertilidade, é a pequena variabilidade ao longo dos vários estágios dos ciclos reprodutivos (ALWARD et al., 2019). Logo, é possível a detecção de animais com alto potencial reprodutivo através da dosagem do AMH de forma precoce.

Em trabalho realizado Grigoletto et al. (2020), os autores encontraram correlação genética positiva e de alta magnitude (0,81 ±0,02), entre a contagem folicular antral após sincronização e níveis de AMH plasmáticos em animais da raça Nelore.

Acredita-se que o AMH pode ser tornar uma ferramenta útil para estimar a eficiência reprodutiva das matrizes, e pode ser usada como ferramenta estratégica para tomar decisões de descarte precoce (ALWARD et al., 2019). Essa avaliação é ideal para aplicação em programas de seleção, voltados para multiplicação de animais com maiores dosagens de

AMH e, consequentemente, com melhores aspectos reprodutivos. Entretanto, ainda são necessárias mais pesquisas para sua consolidação como técnica comercial.

2.5 Eficiência reprodutiva de zebuínos

A eficiência reprodutiva de bovinos de corte é afetada por fatores genéticos, nutricionais e ambientais, além disso é uma característica de extrema importância na produção animal, pois está diretamente ligada a taxa de produção de bezerros e reposição de matrizes em um determinado espaço de tempo em uma propriedade, e consequentemente afetando diretamente sua rentabilidade financeira.

Tanaka (2010) define um animal de boa eficiência reprodutiva, aquele que ainda jovem, 24 meses entra na vida reprodutiva, sendo capaz de se reproduzir em todas as estações de monta, e tendo seus filhos com desempenho superior para características produtivas. A longevidade de uma matriz no rebanho é caracterizada pelo seu tempo de permanência na atividade reprodutiva, sendo tal característica influenciando diretamente o número de bezerros produzidos (MERCADANTE et al., 1996).

Segundo Pereira et al. (2005), a idade média ao primeiro parto de novilhas da raça Tabapuã é de aproximadamente 37 meses, entretanto já em 18 a 20 meses, 60% das novilhas são consideradas aptas para a reprodução (ABCT), tendo uma idade média ao primeiro parto aos 36,8 meses para animais Tabapuã, e 35,8 meses para animais da raça Nelore Mocho (FICHTNER et al., 1988). Já Sereno et al. (2001) observaram a idade ao primeiro parto de aproximadamente aos 40 meses em novilhas Nelore.

Outros índices também são adotados pelos programas de melhoramento genético, como a eficiência reprodutiva (ER), que é responsável por medir a regularidade de partos da vaca. A precocidade, rusticidade e sobrevivência (PRS), que é um índice que combina a precocidade sexual com a capacidade da matriz em levar a gestação a termo e desmamar uma cria viável. A habilidade materna mais provável (HMMP) é o indicador do potencial maternal da vaca em desmamar suas crias em relação a suas contemporâneas e a idade ao primeiro parto (IPP) (PMGZ).

Hoje, ainda, a pecuária Brasileira é vista como heterogênea em termos de tecnologia aplicada a produção de bovinos (CARVALHO e DE ZEN, 2017), sendo essa composta em sua grande maioria por animais de aspectos zebuíno, com amplo nível de mérito genético e eficiência reprodutiva, sofrendo também grande influência de outros fatores como nutrição e sanidade (PEREIRA, 2008).

Portanto, o aperfeiçoamento do manejo reprodutivo e outros aspectos como a nutrição animal, influenciam diretamente a eficiência reprodutiva do rebanho (LOBATO; MAGALHÃES, 2001; VIEIRA et al., 2010). Todavia, a adoção de critérios de seleção para características reprodutivas não deve ser desprezada, visto que animais sexualmente precoces têm maior capacidade produtiva (MERCADANTE et al., 1996), podendo esses, serem utilizados nos programas de melhoramento genético.

2.6 Parâmetros genéticos para características reprodutivas

Os parâmetros genéticos compõem as ferramentas utilizadas para a obtenção de predições das respostas diretas e correlacionadas à seleção, contribuindo na elaboração de índices para selecionar animais (MERCADANTE et al., 1995). Os parâmetros mais utilizados são a herdabilidade e as correlações genéticas. Para obtenção de animais superiores, essa ferramenta se torna muito importante no processo, pois se torna possível direcionar os acasalamentos da melhor forma, obtendo um progresso genético nas características desejadas (LIRA et al., 2008).

O termo herdabilidade (h^2) foi introduzido por Lush (1945), sendo determinada pela razão da variância genética aditiva sobre a variância fenotípica. A herdabilidade pode aumentar se a maior parte da variação fenotípica for de caráter genético, ou diminuir se for de caráter ambiental (LIRA et al., 2008).

Segundo Falconer e Mackay (1996), a herdabilidade é o parâmetro genético de maior relevância, aplicado na determinação do planejamento a ser usado no melhoramento, em razão de medir a capacidade de transmissão desse caráter à sua prole. Assim, uma h^2 alta implica em uma grande variação genética aditiva e uma maior eficiência de ganho genético pela seleção. Quando a herdabilidade é alta, o valor fenotípico é um bom indicativo do valor genético do animal, já para características de baixa herdabilidade, o valor fenotípico não é um bom indicativo do valor genético, pois grande parte da variação da característica é devido a fatores ambientais.

A herdabilidade pode variar em uma escala de 0 a 1 ou de 0 a 100%. De modo geral, valores de herdabilidade de 0 a 20% são considerados baixos, de 20 a 40%, moderados, e acima de 40% altos (CARDELLINO e OSÓRIO, 1999), porém a estimativa de herdabilidade é válida somente para a população analisada no cálculo. A extrapolação para outras populações depende de como se assemelham às estruturas genéticas originais, precisão da medida, condições de meio, dentre outros fatores. Portanto, para se trabalhar com diferentes

rebanhos, deve-se calcular a herdabilidade para cada um deles.

A correlação é o parâmetro que indica o quanto duas características estão associadas geneticamente, capaz de predizer como uma característica deverá se comportar, tendo como parâmetros informações obtidas de outra característica (LIRA et al., 2008). Essa ocorrência é possível devido ao fato de que duas características distintas podem ser controladas por um único gene, denominado pleiotropia. Outro fator que justifica a correlação genética são as ligações gênicas que são genes ligados entre si, ocorrendo assim uma interação que irá determinar uma única característica (LIRA et al., 2008; MARTINS FILHO e LOBO, 1991).

Segundo Falconer e Mackay (1996), as correlações entre duas ou mais características estimam o nível de união entre elas, podendo variar de -1 a 1. De acordo Krapp (2018), os programas de melhoramento genético animal utilizam o conhecimento da correlação genética entre características genéticas de interesse econômico no processo de melhoramento animal, assim permitindo acelerar a seleção de mais de uma característica, influenciando na determinação dos critérios e objetivos de seleção.

A seleção direta para características de fertilidade é tipicamente ineficiente, isso devido ao longo período para avaliação do desempenho das fêmeas e a inconsistência na herdabilidade dessas características (SILVA et al., 2005), devido à alta variação ambiental para tais características (ZINK et al., 2011).

Todavia, a eficiência reprodutiva é um dos fatores determinantes da eficiência total de produção, pois ao selecionar características de precocidade sexual e fertilidade, permitem a tomada de estratégicas como manutenção ou descarte dos animais, com maior pressão de seleção, e consequentemente propiciando maior progresso genético e lucratividade (LOURENÇO, 2011; MELLO et al., 2016; SILVEIRA et al., 2004; VIEIRA et al., 2010). Sendo assim, a busca por características indicadoras de fertilidade pode ser de grande valia, pela identificação de fenótipos que possam ser utilizados na seleção de animais reprodutivamente superiores (MACULAN, 2015).

REFERÊNCIAS

ASSOCIAÇÃO BRASILEIRA DOS CRIADORES DA RAÇA TABAPUÃ (ABCT): CARACTERÍSTICAS DO TABAPUÃ. Uberaba, 2003. Disponível em http://www.tabapua.org.br/index.php/raca-tabapua/caracteristicas-da-raca. Acesso em 09 jun. 2022.

ABIEC - ASSOCIAÇÃO BRASILEIRA DAS INDÚSTRIAS EXPORTADORAS DE CARNE. Beef Report: Perfil da Pecuária no Brasil 2022. 2022. Disponível em: < https://www.abiec.com.br/publicacoes/beef-report-2022/>. Acesso em 15 de março de 2022.

ALWARD, K. J.; BOHLEN, J. F. Overview of Anti-Müllerian hormone (AMH) and association with fertility in female cattle. **Reproduction in Domestic Animals**, v. 55, n. 1, p. 3-10, 2019

BANCSI, L. F. *et al*. Predictors of poor ovarian response in in vitro fertilization: a prospective study comparing basal markers of ovarian reserve. **Fertility and sterility**, v. 77, p. 328-336, 2002.

BATISTA, E. O. S. *et al*. Plasma Antimullerian Hormone as a Predictor of Ovarian Antral Follicular Population in Bos indicus (Nelore) and Bos taurus (Holstein) Heifers. **Reproduction in Domestic Animals**, v. 49, n. 3, p. 448-452, 2014.

BURNS, D. S. *et al*. Numbers of antral follicles during follicular waves in cattle: Evidence for High variation among animals, very high repeatability in individuals and an inverse association with serum follicle-stimulating hormone concentrations. **Biology of Reproduction**, v. 73, n. 1, p. 54-62, 2005.

CARDELLINO, R. A.; OSÓRIO, J. C. S. **Melhoramento Animal para Agronomia, Veterinária e Zootecnia.** 1 ed. Pelotas: Universidade Federal de Pelotas, 1999. 152p.

CARVALHO, T. B.; DE ZEN, S. A cadeia de Pecuária de Corte no Brasil: evolução e tendências. **Revista IPecege**, v. 3, n. 1, p. 85–99, 2017.

COUTINHO, G. T. R. M. *et al.* Avaliação ultrassonográfica da dinâmica folicular e lútea em vacas da raça Guzerá. **Arquivos Brasileiros de Medicina Veterinária e Zootecnia,** Belo Horizonte**.** v. 59, n. 5, p. 1089-1096, 2007.

CUSHMAN, R. A. *et al*. Evaluation of antral follicle count and ovarian morphology in crossbred beef cows: Investigation of influence of stage of the estrous cycle, age, and birth weight. **Journal Animal Science,** Champaign, v.87, n. 6, p.1971–1980, 2009.

EMBRAPA GADO DE CORTE. O Futuro da Cadeia Produtiva da Carne Bovina Brasileira: uma visão para 2040. **Embrapa Gado de Corte-Outras publicações técnicas (INFOTECA-E)**, p. 132, 2020.

FALCONER, D. S.; MACKAY, T. F. C. **Introduction to quantitative genetics**. 4. ed. Edinburgh: Longman, 1996. 464 p.

FICHTNER, S. S.; JARDIM, E. C.; COSTA, J. L. Idade ao primeiro parto de rebanhos das raças tabapuã mocho e nelore mocho. **Anais da Escola de Agronomia e Veterinária**, Goiânia, v. 18, n. 1, p. 121-125, 1988.

GRIGOLETTO, L. *et al*. Genetic Parameters and Genome-Wide Association Studies for Anti-Müllerian Hormone Levels and Antral Follicle Populations Measured After Estrus Synchronization in Nellore Cattle. **Animals**, v. 10, n. 1185, p. 1-15, 2020.

IBGE - INSTITUTO BRASILEIRO DE GEOGRAFIA E ESTATÍSTICA. Produção da Pecuária Municipal 2019. **Pesquisa da Pecuária Municipal**, Rio de Janeiro, v. 49, p.1-12, 2021.

IRELAND, J. L. H. *et al*. Antral Follicle Count Reliably Predicts Number of Morphologically Healthy Oocytes and Follicles in Ovaries of Young Adult Cattle. **Biology of Reproduction**, v. 79, n. 6, p. 1219-1225, 2008.

IRELAND, J. J. *et al.* Does size matter in females? An overview of the impact of the high variation in the ovarian reserve on ovarian function and fertility, utility of anti-Mullerian hormone as a diagnostic marker for fertility and causes of variation in the ovarian reserve in cattle. **Reproduction Fertility and Development**, v. 23, n. 1, p. 1-14, 2010.

IRELAND, J. J. *et al.* Follicle numbers are highly repeatable within individual animals but are inversely correlated with FSH concentrations and the proportion of good-quality embryos after ovarian stimulation in cattle. **Human Reproduction**, v. 22, n. 6, p. 1687-1695, 2007.

KRAPP, A. **Parâmetros genéticos para características de carcaça, peso corporal e idade ao primeiro parto em bovinos da raça Nelore**. 2018. 38 p. Dissertação (Mestrado em Zootecnia) – Instituto de Ciências Agrárias e Ambientais, Universidade Federal de Mato Grosso, Sinop, 2018.

LIRA, T.; ROSA, E. M.; GARNERO, A. D. V. Parâmetros genéticos de características produtivas e reprodutivas em zebuínos de corte. **Ciência Animal Brasileira**, v. 9, n. 1, p. 1-22, 2008.

LOBATO, J. F. P.; MAGALHÃES, F. R. Comportamento reprodutivo de primíparas aos 24 e 36 meses de idade. **Arquivos da faculdade de Veterinária UFRGS,** Porto Alegre. v. 29, n. 2, p. 139- 146, 2001.

LUSH, J. L. Animal Breeding Plans, Ed. 3. Iowa State Univer- sity Press, Ames, IA, 1945.

MACULAN, R. **Características indicadoras de fertilidade em fêmeas da raça Tabapuã**. 2015. 60 p. Dissertação (Mestrado em Zootecnia) - Universidade Federal de Lavras, Lavras, Minas Gerais, 2015.

MACULAN, R. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Animal Reproduction Science**, n. 189, p. 84-92, 2018.

MACULAN, R. **Utilização de características indicadoras de fertilidade na predição do potencial reprodutivo de fêmeas e machos Bos taurus e Bos Indicus**. 2018. 98 p. Tese (Doutorado em Zootecnia) - Universidade Federal de Lavras, Lavras, 2018.

MARTINS FILHO, R.; LÔBO, R.B. Estimates of genetic correlations between sire scrotal circumference and offspring age at first calving in Nellore cattle. **Rev. Bras. Genet.**, v.14, p.209- 212, 1991.

MELLO, R. R. C. *et al*. Parâmetros genéticos de características reprodutivas em bovinos de corte e leite. **Revista Brasileira de Reprodução Animal**, Belo Horizonte, v. 40, n. 2, p. 65-72, 2016.

MERCADANTE, M. E. Z. *et al*. **Estudo genético de características indicadoras da vida útil de fêmeas de um rebanho da raça Nelore**. In: SIMPÓSIO NACIONAL DE MELHORAMENTO ANIMAL, Ribeirão Preto, v. 1, p. 260-262, 1996.

MERCADANTE, M. E. Z.; LÔBO, R. B.; BORJAS A. R. Parámetros genéticos para características de crecimiento em cebuínos de carne. **Archives Latinoamericanos de Production Animal**, v. 3, n. 1, p. 45-89, 1995.

MESQUITA, N. F. *et al*. Vulvar width and rima length as predictors of the ovarian follicular reserve in bovine females. **Journal of Reproduction and Development**, v. 62, n. 6, p. 1-4, 2016.

MODINA, S. *et al*. Reductions in the number of mid-sied antral follicle are associated with markers of premature ovarian senescence in dairy cows. **Reproduction, Fertility and Development**, v. 26, n. 2, p. 235-244, 2014.

MONNIAUX, D. *et al*. Regulation of antiMüllerian hormone production in domestic animals. **Reproduction Fertility and Development**. v. 25, n. 1, p. 1–16, 2012.

MORAIS R. *et al*. Hormônio antimülleriano: pode uma realidade usada em seres humanos ser transferida a bovinos e búfalos. **Revista Brasileira de Reprodução Animal**, Belo Horizonte, v. 41, n. 4, p. 693-698, 2017.

MOROTTI, F. S. *et al*. Is the number of antral follicles an interesting selection criterium for fertility in cattle?. **Animal Reproduction**, v.12, n.3, p.479-486, 2015.

MOROTTI, F. Z. *et al*. Antral follicle count in cattle: advantages, challenges, and controversy. **Animal Reproduction**, v.14, n.3, p.414-420, 2017.

PEREIRA, J. C. C. *et al*. Análise genética de características ponderais e reprodutivas de fêmeas bovinas Tabapuã. **Arquivo Brasileiro de Medicina Veterinária e Zootecnia**, v. 57, n. 2, p. 231-236, 2005.

PEREIRA, J. C. C. **Melhoramento genético aplicado à produção animal**. 5 ed. Belo Horizonte: FEPMVZ, 2008. v. 1, 618 p.

PINTO, T. L. C. **Relações entre população folicular antral, hormônio anti-mulleriano, mastite e fertilidade em bovinos**. 2018. 69 p. Tese (Doutorado em Ciências Veterinárias) - Universidade Federal de Lavras, Lavras, 2018.

RICO, C. *et al*. Anti-Mullerian Hormone Is an Endocrine Marker of Ovarian Gonadotropin Responsive Follicles and Can Help to Predict Superovulatory Responses in the Cow. **Biology of Reproduction**, v. 80, n. 1, p. 50-59, 2009.

RICO, C. *et al.* Regulation of Anti-Müllerian hormone production in the cows: a multiscale study at endocrine, ovarian, follicular, and granulosa cell levels. **Biology of Reproduction**, v, 84, n, 3, p. 560–571, 2011.

RODRIGUES, L. M. S.; MARTA-COSTA, A. A. Competitividade das exportações de carne bovina do Brasil: uma análise das vantagens comparativas. **Revista de Economia e Sociologia Rural**, v. 59, n. 1, p. 1–14, 2021.

SERENO, J. R. B. *et al.* Estimativa de la edad y peso de la primeira monta de novillas em el Pantanal Brasileño. **Pesquisa Agropecuária Brasileira,** Brasília, v. 36, n. 12, p. 1561-1565, 2001.

SILVA, J. A. *et al.* Estudo genético da precocidade sexual de novilhas em um rebanho Nelore. **Revista Brasileira de Zootecnia,** v. 34, n. 5, p. 1568-1572, 2005.

SILVEIRA, L. C. *et al*. Fatores ambientais e parâmetros genéticos para características produtivas e reprodutivas em um rebanho Nelore no estado do Mato Grosso do Sul. **Revista Brasileira de Zootecnia**, v. 33, n. 6, p. 1432-1444, 2004.

TANAKA, A. L. R. **Eficiência reprodutiva de fêmeas Nelore**. 2010. v, 103 f. Tese (doutorado) – Faculdade de Ciências Agrárias e Veterinárias, Universidade Estadual Paulista, Jaboticabal, 2010.

USDA – UNITED STATES DEPARTMENT OF AGRICULTURE FOREIGN AGRICULTURAL SERVICE–. **Production, Supply ans Distribution**. Disponível em: <https://apps.fas.usda.gov/psdonline/app/index.html#/app/advQuery>. Acesso em: 10 março 2022.

VASCONCELOS, G. L. *et al*. Effects of vulvar width and antral follicle count on oocyte quality, in vitroembryo production and pregnancy rate in *Bos taurus taurus* and *Bos taurus indicus* cow. **Animal Reproduction Science** v. 217, n. 106357, 2020.

VIEIRA, D. H. A. *et al.* Non genetics effects on reproductive traits in nellore female. **Revista Brasileira de Medicina Veterinária,** Belo Horizonte, v. 32, n. 2, p. 79-88, 2010.

WALLACE W. H.B. & KELSEY T.W. Human Ovarian Reserve from Conception to the Menopause. **PLOS ONE** 5(1): e8772. https://doi.org/10.1371/journal.pone.0008772, 2010.

ZINK, V. *et al*. Genetic parameters for female fertility, locomotion, body condition score, and linear type traits in Czech Holstein cattle. **Journal of Dairy Science**. v. 94, n. 10, p. 5176-5182, 2011.

SEGUNDA PARTE

ARTICLE 1

Association between vulvar morphometry, antral follicular count and Anti-Mullerian hormone in Tabapuã females

Formatted according to the Livestock Science Journal

G. C. Brilhante[a]; R. Maculan[b]; B. P. G. Araujo[a]; M. M. Baena[c]; J. C. de Souza[a]; S. L. C. Meirelles[a]*

[a] Federal University of Lavras, Department Animal Science, University Campus, postal code 3037, Downtown, CEP 37200000, Lavras/MG, Brazil.
[b] Federal Institute South of Minas Gerais, CEP 37750000, Machado/MG, Brazil.
[c] National Association of Breeders and Researchers, Rua João Godoy, 463 - Jardim America, CEP 14020-230, Ribeirão Preto/SP, Brazil.

*Corresponding author: Federal University of Lavras, Department Animal Science, University Campus, postal code 3037, Downtown, CEP 37203-202, Lavras/MG, Brazil; E-mail: sarah@ufla.br

ABSTRACT

The search for identifying precocious animals has been advancing over the years, making it necessary to produce better animals, but some traits are not easy to select. The response to the selection of reproductive traits is slower, as they generally have low heritability, being greatly influenced by environmental factors. With the aim of accelerating the genetic improvement and selection of these reproductive traits, the search for fertility indicator traits early and easily measured is interesting, as it would facilitate the selection process in the field. The aim of this work was to look for traits that indicate fertility in Zebu females, estimate the heritability and evaluate the genetic correlation between them. Data was collected from 934 females, born between 1993 and 2020, evaluating traits such as vulvar size, antral follicle counts (AFC), concentration of Anti-Mullerian hormone (AMH) of the Tabapuã breed. The genetic parameters of the traits were calculated by Bayesian analysis using executables from the BLUPF90 family, and for traits related to vulvar size and AMH concentration, the executable GIBBSF90 was used, and for AFC, the executable THRGIBBSF90 was used. The analyzes were carried out using a linear mixed model that included the fixed effects of contemporary group formed by year of birth, farm and parity, in addition to the fixed effects of age and weight class and the random effect, the animal itself. In this study, moderate heritability was found for the traits: RIMA (0.33); AFC (0.24) and LVULVA (0.20) showing that they respond to direct selection. There is no genetic correlation between the traits studied.

Keywords: rima length; antral follicle count; genetic correlation; heritability; vulvar width.

1 INTRODUCTION

The demand for animal protein is increasing and, consequently, producers need to become more efficient in livestock, being necessary to select genetically superior individuals. For the identification of superior animals, it is necessary to include the use of genetic evaluations of the cows, planning of directed matings, use of reproductive biotechnologies, being able to execute a greater selection pressure. Direct selection for fertility can become complex, due to the long period of evaluation of female performance and the low heritability of these traits. (ZINK et al., 2011).

The identification of more easily measurable traits for fertility is necessary, so that the cows can be evaluated earlier (SILVA et al., 2005). Some studies indicate that the size of the external genitalia can be used as an indicator of fertility (MESQUITA et al., 2016; MACULAN et al., 2018; VASCONCELOS et al., 2020). Moreover, they have already proven that AGC is a good predictor of follicular reserve and ovarian functionality (GRIGOLETTO et al., 2020; IRELAND et al., 2008; MACULAN el al., 2018; MODINA et al., 2014).

The estimation of the size of the follicular reserve is performed indirectly, based on the antral follicle count (AFC) in the ovaries. However, this technique requires skilled labor and the use of ultrasound. Therefore, finding traits that can predict AFC in a more practical, cheaper and, above all, early way is essential for the process of genetic improvement to be more efficient.

Another parameter used as a fertility indicator is AMH, which is strongly associated with AFC (ALWARD e BOHLEN 2019; IRLANDA et al., 2011; GRIGOLETTO et al., 2020; MACULAN et al., 2018). Its advantage is the small variability over the various stages of the reproductive and estrous cycles of cattle, but with a high measurement cost (BALDRIGHI et al., 2014; BARRUSELLI et al 2015; MACULAN et al., 2018, ALWARD et. al., 2019).

Recent studies (Maculan et al. 2018; Mesquita et al. 2016; Vasconcelos et al. 2020) have shown that AMH concentration and the size of reproductive structures (mainly the vulva) can be phenotypically correlated with AFC. However, Ribeiro et al. (2020) addressed that it has not been widely studied so far whether these traits have a genetic correlation and whether they are responsive to selection.

Understanding the correlations between fertility markers and external phenotypic traits can contribute to indirect selection. In this case, easy-to-measure and low-cost traits can be used as a selection criterion to maximize reproductive efficiency (MACULAN et al., 2018).

This strategy would optimize the selection system, and consequently the producers would achieve a faster return on the investment, in addition to increasing the genetic gain, as it contributes to the reduction of generation interval.

Therefore, the aim of this study was to estimate the genetic parameters between antral follicular count, AMH concentration, vulvar measurements in Tabapuã cattle.

2 MATERIAL E METHODS

2.1 Location and Animals

Data was collected from 934 Tabapuã females aged between two and twenty-one years old. This database was generated in the collections carried out in 2013 and 2021, containing information on reproductive traits from nine farms located in the states of Rio de Janeiro (Fazenda Rodeio Gaúcho, Fazenda São Bento), Minas Gerais (Fazenda Chapadão and Fazenda Quatro Irmãs) and São Paulo (Fazenda Água Milagrosa, Fazenda Porto Seguro, Fazenda Córrego dos Dourados and Fazenda São José das Palmeiras).

The animals were raised in an extensive system and with mineral supplementation. The females had a known father and mother, which came from natural mating, artificial insemination or through embryos. The animals that obtained the measured measurements were not in heat at the time of collection, were not pregnant and were more than 30 days postpartum.

After database consistency, 800 animals were taken for analysis. Females were classified by parity, being divided into heifers n=315 (animals without any calving) and cows n=485 (animals with one or more calvings). All cows had known father and mother.

The zootechnical data file contained the following information on reproductive traits: antral follicle count (AFC), rima length (RL), vulvar width (VW), Anti-Mullerian hormone (AMH) dosage.

2.2 Phenotypic measurements
2.2.1 Antral follicle count

Antral follicle count was performed once in each animal by visualizing the follicles in the antral stage, with a diameter greater than or equal to 3 mm in the right and left ovaries, with the aid of a transrectal ultrasonography model DM10 - DOMED with a 7.0 MHz linear

probe. The sum of the number of follicles from the two ovaries was used to define the total AFC for the analyses. Three classes were created for the AFC analysis, according to the database obtained and their frequencies: low class (less than 26 visible follicles), middle class (between 27 and 44 visible follicles) and upper class (more than 45 follicles visible on ultrasound) (Table 1).

Table 1 - Frequency (N) and percentage according to antral follicle count class - AFC of Tabapuã females.

AFC Class	N	Percentage (%)
Low	252	33.33
Medium	243	32.14
Upper	261	34.52
Total	756	100.00

2.2.2 External genital measurements

Vulvar measurements (rima length and vulvar width) were obtained using a digital caliper, according to the methodology described by Mesquita et al. (2016). The rima length (RIMA) was considered the length between the dorsal and ventral commissures of the vertical slit of the animal's vulva (Figure 1 A), whereas the vulvar width (VW) was obtained between the distances of the sides of the vulva from the midpoint of the rima, at an angle of 90 degrees (Figure 1 B).

Figure 1. Measurements of rima lenght and vulvar width
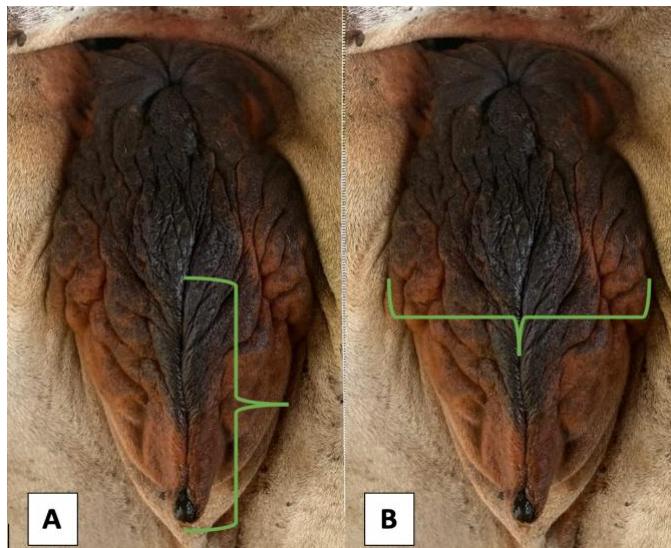
Legend: (A) rima length (A); vulvar width (B).

2.2.3 AMH dosage

AMH quantification was performed in 845 animals, 323 nulliparous, and 522 primiparous and multiparous. Blood samples were collected by coccygeal venipuncture in 10 mL vacuum tubes (BD VACUTAINER®, São Paulo, BR). Serum was obtained after centrifugation at 3,000g for 15 minutes and kept frozen at -20°C for later analysis. AMH serum concentrations were determined by ELISA (Enzyme-Linked Immunosorbent Assay kit; Ansh Labs, Webster, Texas, USA), previously validated for cattle (IRELANDA et al., 2008). One sample was conducted with a sensitivity of 0.011 ng/mL, being intra and inter assay with CV% 2.2%. The assays were carried out at the IgAc laboratory (GENESE Institute of Scientific Investigations, São Paulo, BR).

2.3 Statistical analyzes

The consistency of the database was carried out using the SAS statistical program (SAS, 2010), and after the exploratory analysis, discrepant information was removed (outliers), according to the methodology described by Freitas et al. (2008).

Contemporary groups (CG) were formed by the following variables: year of birth, farm and parity, and those CG that had less than three observations were eliminated. In order to identify which environmental effects significantly influenced (P<0.05) the evaluated traits, analyzes of variance were carried out using the least squares method, using the GLM procedure of the SAS software (SAS, 2010) considering a model with fixed effects of farm, parity, year of birth.

The animal model used for all traits studied included, as systematic effects, the contemporary group, the age class, and the live weight class of the animal at the time of measurements, and as the random effect, the animal itself.

The statistical model used, in matrix form, is described as follows:

$$y = \mathbf{X}\beta + \mathbf{Z}\alpha + e \ (1.0)$$

where:

y = vector of dependent variables;

X = fixed effects incidence matrix;

β = vector of fixed effects;

Z = random effects incidence matrices;

α = vector of direct additive genetic effects;

e = vector of residual random errors associated with observations.

The genetic parameters for vulvar traits (RIMA and VW) and AMH and were estimated through two-trait Bayesian analyses, using GIBBSF90 software (MISZTAL et al., 2002), as they are continuous traits.

For antral follicle count (AFC), the genetic parameters were estimated through two-trait Bayesian analysis, using the THRGIBBSF90 software (Misztal et al., 2002), which allows the study of continuous and categorical traits in the same analysis. For all traits, later estimates were obtained using the executable POSTGIBBSF90 (Misztal et al., 2002). The samples of the (co)variance components were obtained from chains of one million iterations, in which the initial 100,000 iterations were discarded (burn-in) and the samples were removed every 5 cycles (thinning interval).

To perform the convergence tests, the Geweke, Heidel & Ralftery and Raftery & Lewis methods were used using the "BOA" package of the R program (the R v. 3.5.0 (R Core Team, 2018).

3 RESULTS

The mean value observed for AFC and AMH in the present study showed high variation in the population studied, with a coefficient of variation (CV) above 60. The mean value obtained for RIMA and VW did not show a large variation (Table 2).

Table 2 - Descriptive statistics of antral follicle count (AFC), rima length (RIMA), vulvar width (VW), Anti-Mullerian Hormone (AMH), age and weight at the time of measurements of Tabapuã females.

Trait	N	Mean ± SD	Minimum	Maximum	CV
Age (days)	800	2164.7 ± 1489.2	310.0	6127.0	68.3
Weight (Kg)	800	447.7 ± 107.3	192.0	762.0	23.9
AFC (unit)	756	40.9 ± 24.7	2.0	174.0	60.4
RIMA (cm)	800	8.8 ± 1.8	3.3	14.8	21.1
VW (cm)	800	7.6 ± 2.02	3.0	17.0	26.6
AMH (ng/mL)	442	0.8 ± 0.5	0.0	4.5	68.3

N = Number of observations; SD = standard deviation, CV = coefficient of variation.

The weight and age of the animals obtained presented a great variation 447.7±107.3, 2164.7±1489.2 respectively, presenting a CV of 23.9 and 68.3, showing the heterogeneity of the analyzed database (Table 2).

Table 3 – Genetic (above diagonal) and phenotypic correlations (below diagonal) between reproductive traits evaluated in Tabapuã females. Heritability's are diagonally in bold.

	RIMA	VW	AMH	AFC
RIMA	**0.32**	0.50	-0.23	-0.39
VW	0.49**	**0.20**	-0.26	-0.05
AMH	-0.09	-0.07	**0.16**	0.65
AFC	0.10*	0.13*	0.43**	**0.24**

*$P < 0.005$ e **$P < 0.0001$. RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; AFC = antral follicle count.

In this study, moderate herdability were found for the traits: RIMA (0.33); AFC (0.24); LVULVA (0.20) and their respective credibility intervals are represented in the table 4. For these traits, we can say that there is a correlation between the phenotype and the genetic value of the individual, with selection being more responsive.

Table 4 – Additive genetic variance, credible interval, heritability and credible intervals of reproductive traits in Tabapuã females.

Parameters	Vga	CI	h^2	CI h^2
RIMA	0.690	0.310 - 1.080	0.32	0.158 - 0.508
VW	0.392	0.075 -0.719	0.20	0.043 - 0.376
AMH	0.106	0.023 - 0.192	0.16	0.087 - 0.586
AFC	0.484	0.007 - 0.960	0.24	0.008 - 0.484

Vga= mean of additive genetic variance; CI= credibility interval, h^2= herdability, IC h^2= credibility interval of heritability, RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; AFC = antral follicle count.

For traits related to reproductive aspects, a moderate phenotypic correlation was observed between VW and RIMA (0.49) and AFC and AMH (0.43) (Table 3). For the other reproductive traits there was low or no correlation for the present study.

Table 5 – Additive genetic covariance, its credibility intervals and genetic correlations between reproductive traits in Tabapuã females.

Correlation	COVg	CI	GC
RIMA x VW	0.548	0.092 - 1.000	0.50
RIMA x AMH	-0.155	-0.983 – 1.000	-0.23
RIMA x AFC	-0.446	-0.999 – 0.496	-0.39
VW x AMH	-0.271	-0.100 – 0.939	-0.26
VW x AFC	-0.067	-0.999 – 0.943	-0.05
AMH x AFC	0.670	-0.006 – 1.000	0.65

COVg= mean of additive genetic covariance, CI= credibility interval, GC= genetic correlation, RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; AFC = antral follicle count.

Regarding the genetic correlations between the reproductive traits, a moderate correlation was observed between RIMA and VW (0.50). (Table 5). However, by observing the credibility intervals of the other analyses, we can say that there are no genetic correlations between the traits.

4 DISCUSSION

AFC is an important trait to be evaluated to determine reproductive indicators of females (GRIGOLETTO et al., 2020). The mean value observed for the AFC in the present study was 40.99±24.7 with variation from 2 to 174 follicles. These results are in line with Maculan (2015)

who observed a very close value of AFC (38.9±28.00) for Tabapuã breed and with Pinto (2018) who obtained a similar mean of 33.95 ± 4.13 follicles for Girolando breed.

A high variation in the observed antral follicle count was described in the Nellore breed (n=595) by Rodrigues et al. (2015), who reported oscillation from 8 to 145 follicles after synchronization and by Baldrighi et al. (2014), who showed oscillation in the Gir breed (n=10) from 22 to 125 follicles and in the Holstein breed (n=15) heifers, from 18 to 110 follicles after hormonal protocol. On the other hand, Batista et al. (2014), evaluating synchronized Nellore animals (n=16), reported a variation from 18 to 85 follicles and Holstein animals (n=16) a variation from 8 to 51 follicles. These disparities have already been reported and it is believed that they may be due to genetic inequalities, due to the number of individuals analyzed, the feeding of the cows, the type of hormonal protocol used and the great variation of age group in the present study (Sartori et al., 2016; Maculan, 2018).

Mean rima length (8.85±1.8) and vulva width (7.6±2.02) were similar to those found by Maculan et al. (2018) and Ribeiro et al. (2020), with the same breed. It is believed that the similarity is due to the breed studied and the similarity of the age group of the cows between the databases. Variation coefficients were also of low magnitude for these traits.

The variation observed in the concentration of AMH hormone (0.01 to 4.51 ng/mL) was similar to that observed by Maculan et al. (2018) (0.014–4.516 ng/mL), evaluating Tabapuã females. For Nellore heifers, Grigolleto et al. (2020) found a mean of 1.10±0.36, slightly higher than that verified in the present study. According to Batista et al. (2014), values of this hormone may vary between breeds, and Baruselli et al. (2015) stated that its concentration may have a small drop throughout life, which may explain the higher concentration obtained by Grigolleto et al. (2020).

For traits related to reproductive aspects, a moderate phenotypic correlation was observed between VW and RIMA (0.49) and AFC and AMH (0.43) (Table 3). Analyzing 591 Tabapuã females, Maculan et al. (2018), estimated a similar phenotypic correlation between AFC and AMH (0.57), but divergent in relation to the correlation between RIMA and VW (0.09). These results may be due to the database being similar in relation to the origin and breed of the animals. The other phenotypic correlations are not relevant because they are not repeated in the genetic correlations, being traits greatly influenced by the environment.

The heritability obtained for AFC was 0.24, similar to that found by Walsh et al. (2014) in Holstein heifers (0.25) and cows (0.31), and by Grigoletto et al. (2020) who found a heritability of 0.30 for the Nellore breed. Thus, using the AFC as a selection criterion will promote genetic progress, which can be achieved through direct genetic selection.

For the AMH, the heritability found was low (0.16), different from those found by Grigoletto et al. (2020), Gobikrushanth, et al. (2018) and Nawaz et al. (2018) including genomic information which observed a moderate to high heritability, 0.28, 0.36 and 0.46 respectively. This difference in magnitude may be due to variations in terms of genetic values and phenotypes between populations, due to the smaller database used by Grigoletto et al. (2020) (n=289) and by Gobikrushanth et al. (2018) (n=198), It is interesting to carry out further studies to better understand the genetic parameters involving AMH.

In this study, it was verified that the genetic correlation between RIMA and VW (0.50) (Table 5) is positive and moderate, therefore, selecting for the increase of one of them will cause a correlated response for the other trait in the same direction.

However, by observing the credibility intervals of the other analyses, we can say that there are no genetic correlations between the traits. Thus, further studies should be conducted to confirm these relationships between these traits.

Grigoletto et al. (2020) was found a genetic correlation between AMH and AFC for the Nellore breed (0.81). The greater magnitude found may be due to the breed studied and also to the hormonal protocol performed on the cows, ensuring that all were in the same phase of the estrous cycle when measuring the data. These results confirm the importance of measuring AMH concentration early so that superior animals for follicular responses can be identified. However, more studies are needed that take into account the credibility interval of the analyzes so that we can more precisely conclude the genetic correlation between AMH and AFC.

According to the results obtained in this study, we conclude that these characteristics are independent, that is, the selection of one will not cause any correlated response in the other. One of the reasons for this large credibility interval found may be due to the number of data studied. Thus, further research involving these characteristics should be conducted.

5 CONCLUSIONS

In this study, moderate heritabilities was found rima length, antral follicle count and vulvar width, showing that they respond to direct selection. However, it was observed that there is no genetic correlation between the characteristics studied. So further research involving these characteristics should be conducted.

REFERENCES

ALWARD, K. J.; BOHLEN, J. F. Overview of Anti-Müllerian hormone (AMH) and association with fertility in female cattle. **Reproduction in Domestic Animals (Wiley)**, v. 55, n. 1, p. 3-10, 2019

BALDRIGHI, J. M. *et al*. Anti-Müllerian hormone concentration and antral ovarian follicle population in murrah heifers compared to holstein and gyr kept under the same management. **Reproduction in Domestic Animals**, v. 49, n. 6, p. 1015–1020, 2014.

BARUSELLI, P. S. *et al*. Relationship between follicle population, AMH concentration and fertility in cattle. **Animal Reproduction**, v. 12, n. 3, p. 487-497, 2015.

BATISTA, E. O. S. *et al*. Plasma Antimullerian Hormone as a Predictor of Ovarian Antral Follicular Population in *Bos indicus* (Nelore) and *Bos taurus* (Holstein) Heifers. **Reproduction in Domestic Animals**, v. 49, n. 3, p. 448-452, 2014.

FREITAS, A. R. et al. Técnicas de análises exploratórias em dados de cultivares de alfafa. **Revista Brasileira de Zootecnia**, Viçosa, MG, v. 37, n. 9, p. 1532- 1532, 2008.

GRIGOLETTO, L. *et al*. Genetic Parameters and Genome-Wide Association Studies for Anti-Müllerian Hormone Levels and Antral Follicle Populations Measured After Estrus Synchronization in Nellore Cattle, 2020.

GOBIKRUSHANTH M., *et al*. The relationship between serum anti-Müllerian hormone concentrations and fertility, and genome-wide associations for anti-Müllerian hormone in Holstein cows. **Journal of Dairy Science** v. 101, n. 8, p. 7563-7574, 2018.

GRIGOLETTO, L. *et al*. Genetic Parameters and Genome-Wide Association Studies for Anti-Müllerian Hormone Levels and Antral Follicle Populations Measured After Estrus Synchronization in Nellore Cattle. **Animals**, v. 10, n. 1185, p. 1-15, 2020.

IRELAND, J. L. H. *et al*. Antral Follicle Count Reliably Predicts Number of Morphologically Healthy Oocytes and Follicles in Ovaries of Young Adult Cattle. **Biology of Reproduction**, v. 79, n. 6, p. 1219-1225, 2008.

IRELAND, J.J. *et al*. Does size matter in females? An overview of the impact of the high variation in the ovarian reserve on ovarian function and fertility, utility of anti-Mullerian hormone as a diagnostic marker for fertility and causes of variation in the ovarian reserve in cattle. **Reproduction Fertility and Development**, v. 23, p. 1–14, 2011.

MACULAN, R. **Características indicadoras de fertilidade em fêmeas da raça Tabapuã**. 2015. 60 p. Dissertação (Mestrado em Zootecnia) – Universidade Federal de Lavras, Lavras, 2015.

MACULAN, R. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Animal Reproduction Science**, n. 189, p. 84-

92, 2018.

MACULAN, R. **Utilização de características indicadoras de fertilidade na predição do potencial reprodutivo de fêmeas e machos Bos taurus e Bos Indicus**. 2018. 98 p. Tese (Doutorado em Zootecnia) - Universidade Federal de Lavras, Lavras, 2018.

MESQUITA, N. F. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Journal of Reproduction and Development**, v. 62, n. 6, p. 1-4, 2016.

MODINA, S. *et al*. Reductions in the number of mid-sied antral follicle are associated with markers of premature ovarian senescence in dairy cows. **Reproduction, Fertility and Development**, v. 26, n. 2, p. 235-244, 2014.

NAWAZ M. Y. *et al*. Genomic heritability and genome-wide association analysis of anti-Müllerian hormone in Holstein dairy heifers. **Journal of Dairy Science,** v. 101, n. 9, p. 8063-8075, 2018.

PINTO, T. L. C. **Relações entre população folicular antral, hormônio anti-mulleriano, mastite e fertilidade em bovinos**. 2018. 69 p. Tese (Doutorado em Ciências Veterinárias) - Universidade Federal de Lavras, Lavras, 2018.

RIBEIRO, Y. P. **Parâmetros genéticos para características indicadoras de fertilidade em fêmeas da raça Tabapuã**. 2020. 28 p. Trabalho de conclusão de curso (Bacharelado em Zootecnia) - Universidade Federal de Lavras, Lavras, 2020.

RODRIGUES, A. S. *et al*. Antral follicle count in nelore females subjected to fixed-time artificial insemination. **Ciência Rural**, v. 45, n. 4, p. 711-717, 2015.

SARTORI, R. *et al.* Metabolic and endocrine differences between *Bos taurus* and *Bos indicus* females that impact the interaction of nutrition with reproduction. **Theriogenology,** v.86, n.1, p. 32

SILVA, J. A. *et al*. Estudo genético da precocidade sexual de novilhas em um rebanho Nelore. **Revista Brasileira de Zootecnia,** v. 34, n. 5, p. 1568-1572, 2005.

VASCONCELOS, G. L. *et al*. Effects of vulvar width and antral follicle count on oocyte quality, in vitroembryo production and pregnancy rate in Bos taurus taurus and Bos taurus indicus cow. **Animal Reproduction Science,** v. 217, n. 106357, 2020.

ZINK, V. *et al*. Genetic parameters for female fertility, locomotion, body condition score, and linear type traits in Czech Holstein cattle. **Journal Dairy Science**, v. 94, n. 10, p. 5176- 5182, 2011.v. 94, p. 5176- 5182, 2011.

ARTICLE 2

Association between vulvar morphometry, Anti-Mullerian hormone concentration and biometric measurements in Tabapuã females

Formatted according to the Livetock Science Journal

G. C. Brilhante[a]; R. Maculan[b]; B. P. G. Araujo[a]; M. M. Baena[c]; J. C. de Souza[a]; S. L. C. Meirelles[a]*

[a] Federal University of Lavras, Department Animal Science, University Campus, postal code 3037, Downtown, CEP 37200000, Lavras/MG, Brazil.
[b] Federal Institute South of Minas Gerais, CEP 37750000, Machado/MG, Brazil.
[c] National Association of Breeders and Researchers, Rua João Godoy, 463 - Jardim America, CEP 14020-230, Ribeirão Preto/SP, Brazil.

*Corresponding author: Federal University of Lavras, Department Animal Science, University Campus, postal code 3037, Downtown, CEP 37203-202, Lavras/MG, Brazil; E-mail: sarah@ufla.br

ABSTRACT

The search for identifying precocious animals has been advancing over the years, making it necessary to produce better animals, but some traits are not easy to select. The response to the selection of reproductive traits is slower, as they generally have low heritability, being greatly influenced by environmental factors. With the aim of accelerating the genetic improvement and selection of these reproductive traits, the search for fertility indicator traits early and easily measured is interesting, as it would facilitate the selection process in the field. The objective of this work was to look for traits that indicate fertility in Zebu females, estimate the heritability and evaluate the genetic correlation between them. The aim of this work was to look for traits that indicate fertility in Zebu females, estimate the heritability and evaluate the genetic correlation between them. We collected data from 934 females, born between 1993 and 2020, evaluating traits such as vulvar size, concentration of Anti-Mullerian hormone (AMH) and biometric measurements of the Tabapuã breed. The genetic parameters of the traits were calculated by Bayesian analysis using executables from the BLUPF90 family, and for traits related to vulvar size, biometric measurements and AMH concentration, the executable GIBBSF90. The analyzes were carried out using a linear mixed model that included the fixed effects of contemporary group formed by year of birth, farm and parity, in addition to the fixed effects of age and weight class and the random effect, the animal itself. In this study, moderate heritability was found for the characteristics: hearth girth (HG) (0.37), anterior height (AH) (0.42), RIMA (0.32), vulva width (VW) (0.20) showing that they respond to direct selection. There is no genetic correlation between the traits studied.

Keywords: AMH; genetic correlation; heritability; rima lengtht; vulva width.

1 INTRODUCTION

The demand for animal protein is increasing and, consequently, producers need to become more efficient in livestock, being necessary to select genetically superior individuals. For the identification of superior animals, it is necessary to include the use of genetic evaluations of the cows, planning of directed matings, use of reproductive biotechnologies, being able to execute a greater selection pressure. Direct selection for fertility can become complex, due to the long period of evaluation of female performance and the low heritability of these traits (ZINK et al., 2011).

The identification of more easily measurable traits for fertility is necessary, so that the cows can be evaluated earlier (SILVA et al., 2005). Some studies indicate that the size of the external genitalia can be used as an indicator of fertility (MESQUITA et al., 2016; MACULAN et al., 2018; VASCONCELOS et al., 2020). Moreover, they have already proven that AGC is a good predictor of follicular reserve and ovarian functionality (GRIGOLETTO et al., 2020; IRELAND et al., 2008; MACULAN el al., 2018; MODINA et al., 2014).

Another parameter used as a fertility indicator is AMH, which is strongly associated with AFC (ALWARD e BOHLEN 2019; IRLANDA et al., 2011; GRIGOLETTO et al., 2020; MACULAN et al., 2018). Its advantage is the small variability over the various stages of the reproductive and estrous cycles of cattle, but with a high measurement cost (BALDRIGHI et al., 2014; BARRUSELLI et al 2015; MACULAN et al., 2018, ALWARD et. al., 2019).

Recent studies (MACULAN et al. 2018; MESQUITA et al. 2016; VASCONCELOS et al. 2020) have shown that AMH concentration and the size of reproductive structures (mainly the vulva) can be phenotypically correlated with AFC. However, Ribeiro et al. (2020) addressed that it has not been widely studied so far whether these traits have a genetic correlation and whether they are responsive to selection.

Understanding the correlations between fertility markers and external phenotypic traits can contribute to indirect selection. In this case, easy-to-measure and low-cost traits can be used as a selection criterion to maximize reproductive efficiency (MACULAN et al., 2018). This strategy would optimize the selection system, and consequently the producers would achieve a faster return on the investment, in addition to increasing the genetic gain, as it contributes to the reduction of generation interval.

Therefore, the aim of this study was to estimate the genetic parameters between AMH concentration, vulvar measurements and biometric measurements in Tabapuã cattle.

2 MATERIAL E METHODS

2.1 Location and Animals

Data was collected from 934 Tabapuã females aged between two and twenty-one years old. This database was generated in the collections carried out in 2013 and 2021, containing information on reproductive traits from nine farms located in the states of Rio de Janeiro (Fazenda Rodeio Gaúcho, Fazenda São Bento), Minas Gerais (Fazenda Chapadão and Fazenda Quatro Irmãs) and São Paulo (Fazenda Água Milagrosa, Fazenda Porto Seguro, Fazenda Córrego dos Dourados and Fazenda São José das Palmeiras).

The animals were raised in an extensive system and with mineral supplementation. The females had a known father and mother, which came from natural mating, artificial insemination or through embryos. The animals that obtained the measured measurements were not in heat at the time of collection, were not pregnant and were more than 30 days postpartum.

After database consistency, 800 animals were taken for analysis. Females were classified by parity, being divided into heifers n=315 (animals without any calving) and cows n=485 (animals with one or more calvings). All cows had known father and mother.

The zootechnical data file contained the following information on reproductive traits: rima length (RL), vulvar width (VW), Anti-Mullerian hormone (AMH) dosage, biometric measurements, such as: anterior height (AH), rump height (RH), hearth girth (HG), body length (BL), rump length (RL), ileum width (IW), pin width (PW), pelvic depth (PD), thoracic depth (TD), sternum to ground distance (SG).

2.2 Phenotypic measurements
2.2.1 External genital measurements

Vulvar measurements (rima length and vulvar width) were obtained using a digital caliper, according to the methodology described by Mesquita et al. (2016). The rima length (RIMA) was considered the length between the dorsal and ventral commissures of the vertical slit of the animal's vulva (Figure 1 A), whereas the vulvar width (VW) was obtained between the distances of the sides of the vulva from the midpoint of the rima, at an angle of 90 degrees (Figure 1 B).

Figure 1. Measurements of rima lenght and vulvar width
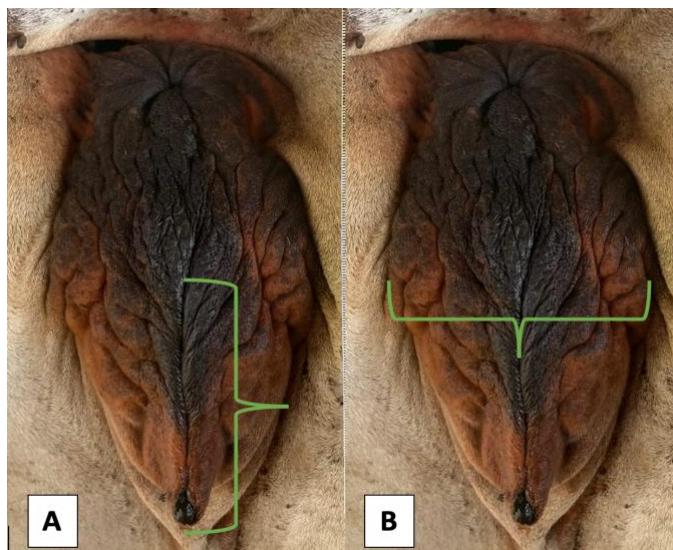
Legend: (A) rima length (A); vulvar width (B).

2.2.3 Biometric measurements

The following body biometric measurements were performed using a hypometer: body length (BL) - distance between the tip of the shoulder and the ischial tuberosity; rump length (RL) - distance between ischium and ileum; thoracic depth (TD) - distance between the withers and the sternum; pelvic depth (PD) - distance between the sacral tuberosity and the insertion of the gastrocnemius muscle; anterior height (AH) - distance between the withers and the ground; rump height (RH) - distance between the sacral tuberosity and the ground; rump width between ischia (PW) - distance between ischial tuberosities; rump width between the ileums (IW) - distance between the iliac tuberosities; hearth girth (HG) - obtained with tape measure, and body weight using an electronic scale attached to the containment chute. The sternum to ground distance (SG) was calculated by subtracting the anterior height from the thoracic depth.

2.2.4 AMH dosage

AMH quantification was performed in 845 animals, 323 nulliparous, and 522 primiparous and multiparous. Blood samples were collected by coccygeal venipuncture in 10 mL vacuum tubes (BD VACUTAINER®, São Paulo, BR). Serum was obtained after centrifugation at 3,000g for 15 minutes and kept frozen at -20°C for later analysis. AMH serum concentrations were determined by ELISA (Enzyme-Linked Immunosorbent Assay kit; Ansh Labs, Webster, Texas, USA), previously validated for cattle (IRELANDA et al., 2008). One sample was conducted with a sensitivity of 0.011 ng/mL, being intra and inter

assay with CV% 2.2%. The assays were carried out at the IgAc laboratory (GENESE Institute of Scientific Investigations, São Paulo, BR).

2.3 Statistical analyzes

The consistency of the database was carried out using the SAS statistical program (SAS, 2010), and after the exploratory analysis, discrepant information was removed (outliers), according to the methodology described by Freitas et al. (2008).

Contemporary groups (CG) were formed by the following variables: year of birth, farm and parity, and those CG that had less than three observations were eliminated. In order to identify which environmental effects significantly influenced (P<0.05) the evaluated traits, analyzes of variance were carried out using the least squares method, using the GLM procedure of the SAS software (SAS, 2010) considering a model with fixed effects of farm, parity, year of birth.

The animal model used for all traits studied included, as systematic effects, the contemporary group, the age class, and the live weight class of the animal at the time of measurements, and as the random effect, the animal itself.

The statistical model used, in matrix form, is described as follows:

$$y = \mathbf{X}\beta + \mathbf{Z}\alpha + e \ (1.0)$$

where:

y = vector of dependent variables;

X = fixed effects incidence matrix;

β = vector of fixed effects;

Z = random effects incidence matrices;

α = vector of direct additive genetic effects;

e = vector of residual random errors associated with observations.

The genetic parameters for vulvar traits (RIMA and VW), AMH and biometric measurements were estimated through two-trait Bayesian analyses, GIBBSF90 software (MISZTAL et al., 2002), as they are continuous traits.

For all traits, later estimates were obtained using the executable POSTGIBBSF90 (Misztal et al., 2002). The samples of the (co)variance components were obtained from chains

of one million iterations, in which the initial 100,000 iterations were discarded (burn-in) and the samples were removed every 5 cycles (thinning interval).

To perform the convergence tests, the Geweke, Heidel & Ralftery and Raftery & Lewis methods were used using the "BOA" package of the R program (the R v. 3.5.0 (R Core Team, 2018).

3 RESULTS

The mean value observed for AMH in the present study showed high variation in the population studied, with a coefficient of variation (CV) above 60. The mean value obtained for RIMA and VW did not show a large variation (Table 1).

Table 1 - Descriptive statistics of antral follicle count (AFC), rima length (RIMA), vulvar width (VW), Anti-Mullerian Hormone (AMH), age and weight at the time of measurements of Tabapuã females.

Trait	N	Mean ± SD	Minimum	Maximum	CV
Age (days)	800	2164.7 ± 1489.2	310.0	6127.0	68.3
Weight (Kg)	800	447.7 ± 107.3	192.0	762.0	23.9
RIMA (cm)	800	8.8 ± 1.8	3.3	14.8	21.1
VW (cm)	800	7.6 ± 2.02	3.0	17.0	26.6
AMH (ng/mL)	442	0.8 ± 0.5	0.0	4.5	68.3

N = Number of observations; SD = standard deviation, CV = coefficient of variation.

The weight and age of the animals obtained presented a great variation 447.7±107.3, 2164.7±1489.2 respectively, presenting a CV of 23.9 and 68.3, showing the heterogeneity of the analyzed database (Table 1).

Table 2– Descriptive statistics of biometric measurements of Tabapuã females.

Trait*	N	Mean ± SD	Minimum	Maximum	CV
BL (cm)	800	143.5 ± 11.9	115.0	186.0	8.3
RL (cm)	800	46.6 ± 4.9	32.0	63.0	10.5
TD (cm)	800	67.4 ± 6.1	48.0	86.0	9.0
PD (cm)	800	63.5 ± 6.9	43.0	86.0	10.9
AH (cm)	800	137.0 ± 7.8	115.0	167.0	5.7
RH (cm)	800	144.3 ± 7.9	125.0	179.0	5.4
PW (cm)	800	21.1 ± 5.0	11.0	39.0	23.8
IW (cm)	800	46.9 ± 6.2	32.0	69.0	13.3
HG (cm)	701	182.2 ± 16.4	111.0	223.0	9.0
SG (cm)	800	69.5 ± 5.9	56.0	100.0	8.5

N = Number of observations; SD = standard deviation; CV = coefficient of variation; BL = body length; RL = rump length; TD = thoracic depth; PD = pelvic depth; FL = forelimb length; RH = rump height; PW = Rump width between the ischium; IW = rump width between the ileums; HG = hearth girth; SG = sternum to ground distance.

Table 3 - Additive genetic variance, credibility interval, heritabilities and credibility intervals of reproductive traits and biometric measurements in Tabapuã females.

Parameters	Vga	CI	h^2	CI h^2
RIMA	0.690	0.310 - 1.080	0.32	0.158 - 0.508
VW	0.392	0.075 -0.719	0.20	0.043 - 0.376
AMH	0.106	0.023 - 0.192	0.16	0.087 - 0.586
BL	9.422	1.395 - 17.630	0.18	0.033 - 0.390
RL	1.137	0.062 - 2.235	0.14	0.056 – 0.604
TD	3.454	0.192 - 7.568	0.37	0.010 - 0.387
PD	2.556	0.206 - 5.140	0.15	0.0001 – 0.257
AH	8.675	4.949 - 12.500	0.42	0.277 - 0.642
RH	8.797	2.674 - 15.280	0.34	0.115 - 0.602
PW	1.201	0.256 - 2.274	0.13	0.016 - 0.242
IW	2.055	0.370 - 3.730	0.23	0.037 - 0.385
HG	5.847	0.378 - 12.410	0.13	0.005 - 0.182
SG	0.418	0.073 -9.649	0.20	0.006 – 0.367

Vga= mean of additive genetic variance; CI= credibility interval, h^2= herdability, IC h^2= credibility interval of heritability, RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; RH = rump height; BL = body length; SG = sternum to ground distance; RL = rump length; TD = thoracic depth; FL = forelimb length; PD = pelvic depth; AH = anterior height; PW = Rump width between the ischium; IW = rump width between the

ileums; HG= hearth girth;

High heritability values were found for the trait: AH (0.42) and moderate values for: RIMA (0.33); VW (0.20); TD (0.37); RH (0.34); IW (0.23); SZ (0.20) and their respective credibility intervals are represented in the table 3. For these traits, we can say that there is a correlation between the phenotype and the genetic value of the individual, with selection being more responsive.

Table 4 – Additive genetic covariance, its credibility intervals and genetic correlations between reproductive traits in Tabapuã females.

Correlations	COVg	CI	GC
RIMA x SG	-0.058	-0.782 – 0.595	-0.05
RIMA x VW	0.548	0.092 - 1.000	0.50
RIMA x AMH	-0.155	-0.983 – 1.000	-0.23
RIMA x BL	0.075	-0.516 – 0.999	0.08
RIMA x RL	0.214	-0.648 – 0.999	0.21
RIMA x TD	0.397	0.032 - 0.999	0.37
RIMA x PD	0.500	-0.100 – 1.000	0.46
RIMA x AH	0.209	-0.173 – 0.599	0.20
RIMA x RH	0.476	0.091 - 0.999	0.45
RIMA x PW	0.227	-0.488 – 0.999	0.21
RIMA x IW	0.167	-0.349 – 0.999	0.13
RIMA x HG	0.016	-0.980 – 1.000	0.01
VW x AMH	-0.271	-0.100 – 0.939	-0.26
VW x BL	-0.153	-0.998 – 0.490	-0.12
VW x RL	-0.446	-1.000 – 0.320	-0.39
VW x TD	-0.095	-0.838 – 0.496	-0.07
VW x PD	-0.043	-0.998 – 0.746	-0.02
VW x AH	0.013	-0.581 – 0.550	0.01
VW x RH	0.225	-0.334 – 1.000	0.21
VW x PW	-0.585	-1.000 – 0.041	-0.51
VW x IW	-0.453	-0.999 – 0.124	-0.39
VW x HG	-0.006	-0.987 – 1.000	-0.01
VW x SG	0.142	-0.538 - 1.000	0.12
AMH x BL	0.326	-0.961 – 1.000	0.36
AMH x RL	0.321	-0.973 – 0.999	0.37
AMH x TD	-0.125	-1.000 – 0.955	-0.09
AMH x PD	-0.312	-1.000 – 0.925	-0.29
AMH x AH	0.044	-1.000 – 0.977	0.08
AMH x RH	0.099	-1.000 – 0.982	0.18
AMH x PW	-0.148	-1.000 – 0.943	-0.14
AMH x IW	0.069	-1.000 – 0.972	0.13
AMH x HG	0.696	-0.078 – 1.000	0.67
AMH x SG	-0.005	-1.000 – 0.976	0.07

COVg= mean of additive genetic covariance, CI= credibility interval, GC= genetic correlation, RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; RH = rump height; BL = body length; SG = sternum to ground distance; RL = rump length; TD = thoracic depth; FL = forelimb length; PD = pelvic depth; PW = Rump width between the ischium; IW = rump width between the ileums; HG= hearth girth.

With regard to genetic correlations of reproductive traits, a moderate correlation was observed between RIMA and VW (0.50), so select for greater RIMA, you will be selecting for greater VW.

However, when looking at the credibility intervals of the other analyses, we can say that there are no genetic correlations between the traits (Table 4). Thus, further studies should be conducted to confirm these relationships between these traits.

4 DISCUSSION

Mean rima length (8.85±1.8) and vulva width (7.6±2.02) were similar to those found by Maculan et al. (2018) and Ribeiro et al. (2020), with the same breed. It is believed that the similarity is due to the breed studied and the similarity of the age group of the cows between the databases. Variation coefficients were also of low magnitude for these traits.

The variation observed in the concentration of AMH hormone (0.01 to 4.51 ng/mL) was similar to that observed by Maculan et al. (2018) (0.014–4.516 ng/mL), evaluating Tabapuã females. For Nellore heifers, Grigolleto et al. (2020) found a mean of 1.10±0.36, slightly higher than that verified in the present study. According to Batista et al. (2014), values of this hormone may vary between breeds, and Baruselli et al. (2015) stated that its concentration may have a small drop throughout life, which may explain the higher concentration obtained by Grigolleto et al. (2020).

Regarding the descriptive statistics for the biometric measurements of Tabapuã females, Mourão et al. (1999) describe that in Zebu cows (n=183) the mean of TD is 76.11±4.15 and IW is 50.18±2.77. These values described for PW do not differ from those found in the present study. The TD is above the one found in this study, which may be due to the breed or even to the evolution of the Zebu herd in recent years. More studies are needed to better understand biometric measurements to contribute to the genetic improvement of breeds (ECKSTEIN et al., 2021).

There was a great variation for some traits, such as live weight, age, BL and HG in the studied population. It can be attributed to the great diversity of animals evaluated, including animals of different ages from nine different properties.

The heritability obtained for AMH found was low (0.16), different from those found by Grigoletto et al. (2020), Gobikrushanth, et al. (2018) and Nawaz et al. (2018) including genomic information which observed a moderate to high heritability, 0.28, 0.36 and 0.46 respectively. This difference in magnitude may be due to variations in terms of genetic values and phenotypes between populations, due to the smaller database used by Grigoletto et al.

(2020) (n=289) and by Gobikrushanth et al. (2018) (n=198). It is interesting to carry out further studies to better understand the genetic parameters involving AMH.

In this study, it was verified that the genetic correlation between RIMA and VW (0.50) is positive and moderate, therefore, selecting for the increase of one of them will cause a correlated response for the other trait in the same direction.

However, when looking at the credibility intervals of the other analyses, we can say that there are no genetic correlations between the traits (Table 5). Thus, further studies should be conducted to confirm these relationships between these traits.

Lima et al. (2013) observed in Nelore bulls high genetic correlations observing visual score between sexuality with body structure (0.90), but more studies to know if the size of the external genitalia is indicated to select traits of body structure are still needed but the credibility interval was not used to estimate these values.

According to the results obtained in this study, we conclude that these traits are independent, that is, the selection of one will not cause any correlated response in the other. One of the reasons for this large credibility interval found may be due to the number of data studied. Thus, further research involving these traits should be conducted.

5 CONCLUSIONS

In this study, moderate heritabilities was found for rima length, thoracic hearth girth, anterior height and vulvar width, showing that they respond to direct selection. However, it was observed that there is no genetic correlation between the traits studied. So further research involving these traits should be conducted.

REFERENCES

ALWARD, K. J.; BOHLEN, J. F. Overview of Anti-Müllerian hormone (AMH) and association with fertility in female cattle. **Reproduction in Domestic Animals (Wiley)**, v. 55, n. 1, p. 3-10, 2019

BALDRIGHI, J. M. *et al*. Anti-Müllerian hormone concentration and antral ovarian follicle population in murrah heifers compared to holstein and gyr kept under the same management. **Reproduction in Domestic Animals**, v. 49, n. 6, p. 1015–1020, 2014.

BARUSELLI, P. S. *et al*. Relationship between follicle population, AMH concentration and fertility in cattle. **Animal Reproduction**, v. 12, n. 3, p. 487-497, 2015.

BATISTA, E. O. S. *et al*. Plasma Antimullerian Hormone as a Predictor of Ovarian Antral Follicular Population in *Bos indicus* (Nelore) and *Bos taurus* (Holstein) Heifers. **Reproduction in Domestic Animals**, v. 49, n. 3, p. 448-452, 2014.

ECKSTEIN, M. J. *et al*. Avaliação do desempenho produtivo e reprodutivo, biometria corporal e genealogia de bovinos das raças zebuínas e suas correlações. **Anais da Jornada de Iniciação Científica e Tecnológica**, 2021, Realeza, v. 1, n. 11, 2021. p. 1-4.

FREITAS, A. R. et al. Técnicas de análises exploratórias em dados de cultivares de alfafa. **Revista Brasileira de Zootecnia**, Viçosa, MG, v. 37, n. 9, p. 1532- 1532, 2008.

GOBIKRUSHANTH M., *et al*. The relationship between serum anti-Müllerian hormone concentrations and fertility, and genome-wide associations for anti-Müllerian hormone in Holstein cows. **Journal of Dairy Science** v. 101, n. 8, p. 7563-7574, 2018.

GRIGOLETTO, L. *et al*. Genetic Parameters and Genome-Wide Association Studies for Anti-Müllerian Hormone Levels and Antral Follicle Populations Measured After Estrus Synchronization in Nellore Cattle. **Animals**, v. 10, n. 1185, p. 1-15, 2020.

IRELAND, J. L. H. *et al*. Antral Follicle Count Reliably Predicts Number of Morphologically Healthy Oocytes and Follicles in Ovaries of Young Adult Cattle. **Biology of Reproduction**, v. 79, n. 6, p. 1219-1225, 2008.

IRELAND, J.J. *et al*. Does size matter in females? An overview of the impact of the high variation in the ovarian reserve on ovarian function and fertility, utility of anti-Mullerian hormone as a diagnostic marker for fertility and causes of variation in the ovarian reserve in cattle. **Reproduction Fertility and Development**, v. 23, p. 1–14, 2011.

LIMA, P.R.M. *et al*. Genetic parameters for type classification of Nelore cattle on central performance tests at pasture in Brazil, **Trop Anim Health Prod** 45, 1627–1634, 2013.

MACULAN, R. **Características indicadoras de fertilidade em fêmeas da raça Tabapuã**. 2015. 60 p. Dissertação (Mestrado em Zootecnia) – Universidade Federal de Lavras, Lavras, 2015.

MACULAN, R. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Animal Reproduction Science**, n. 189, p. 84-92, 2018.

MACULAN, R. **Utilização de características indicadoras de fertilidade na predição do potencial reprodutivo de fêmeas e machos Bos taurus e Bos Indicus**. 2018. 98 p. Tese (Doutorado em Zootecnia) - Universidade Federal de Lavras, Lavras, 2018.

MESQUITA, N. F. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Journal of Reproduction and Development**, v. 62, n. 6, p. 1-4, 2016.

MODINA, S. *et al*. Reductions in the number of mid-sied antral follicle are associated with markers of premature ovarian senescence in dairy cows. **Reproduction, Fertility and Development**, v. 26, n. 2, p. 235-244, 2014.

MOURÃO, G. B. *et al*. Diferenças genéticas e estimação de coeficientes de herdabilidade para características morfológicas em fêmeas zebus e F1 holandês x zebu. **Revista Brasileira de Zootecnia**, v.28, n.1, p. 44-54, 1999.

NAWAZ M. Y. *et al*. Genomic heritability and genome-wide association analysis of anti-Müllerian hormone in Holstein dairy heifers. **Journal of Dairy Science,** v. 101, n. 9, p. 8063-8075, 2018.

RIBEIRO, Y. P. **Parâmetros genéticos para características indicadoras de fertilidade em fêmeas da raça Tabapuã**. 2020. 28 p. Trabalho de conclusão de curso (Bacharelado em Zootecnia) - Universidade Federal de Lavras, Lavras, 2020.

SILVA, J. A. *et al*. Estudo genético da precocidade sexual de novilhas em um rebanho Nelore. **Revista Brasileira de Zootecnia,** v. 34, n. 5, p. 1568-1572, 2005.

VASCONCELOS, G. L. *et al*. Effects of vulvar width and antral follicle count on oocyte quality, in vitroembryo production and pregnancy rate in Bos taurus taurus and Bos taurus indicus cow. **Animal Reproduction Science,** v. 217, n. 106357, 2020.

ZINK, V. *et al*. Genetic parameters for female fertility, locomotion, body condition score, and linear type traits in Czech Holstein cattle. **Journal Dairy Science**, v. 94, n. 10, p. 5176- 5182, 2011.v. 94, p. 5176- 5182, 2011.

ARTICLE 3

Association between vulvar morphometry, Anti-Mullerian hormone concentration and reproductive indices for PMGZ in Tabapuã females

Formatted according to the Livetock Science Journal

G. C. Brilhante[a]; R. Maculan[b]; B. P. G. Araujo[a]; M. M. Baena[c]; J. C. de Souza[a]; S. L. C. Meirelles[a]*

[a] Federal University of Lavras, Department Animal Science, University Campus, postal code 3037, Downtown, CEP 37200000, Lavras/MG, Brazil.
[b] Federal Institute South of Minas Gerais, CEP 37750000, Machado/MG, Brazil.
[c] National Association of Breeders and Researchers, Rua João Godoy, 463 - Jardim America, CEP 14020-230, Ribeirão Preto/SP, Brazil.

*Corresponding author: Federal University of Lavras, Department Animal Science, University Campus, postal code 3037, Downtown, CEP 37203-202, Lavras/MG, Brazil; E-mail: sarah@ufla.br

ABSTRACT

The search for identifying precocious animals has been advancing over the years, making it necessary to produce better animals, but some traits are not easy to select. The response to the selection of reproductive traits is slower, as they generally have low heritability, being greatly influenced by environmental factors. With the aim of accelerating the genetic improvement and selection of these reproductive traits, the search for fertility indicator traits early and easily measured is interesting, as it would facilitate the selection process in the field. The aim of this work was to look for traits that indicate fertility in Zebu females, estimate the heritability and evaluate the genetic correlation between them. We collected data from 934 females, born between 1993 and 2020, evaluating traits such as vulvar size, concentration of Anti-Mullerian hormone (AMH) and reproductive indices of the breeding program of Zebu cattle (PMGZ) of the Tabapuã breed. The genetic parameters of the traits were calculated by Bayesian analysis using executables from the BLUPF90 family, and for traits related to vulvar size, reproductive indices and AMH concentration, the executable GIBBSF90. The analyzes were carried out using a linear mixed model that included the fixed effects of contemporary group formed by year of birth, farm and parity, in addition to the fixed effects of age and weight class and the random effect, the animal itself. In this study, moderate heritability was found for the traits: RIMA (0.32), vulva width (VW) (0.20), age at first calving (AAFC) (0.28) and most probable producing ability (MPPA) (0.88) showing that they respond to direct selection. There is no genetic correlation between the traits studied.

Keywords: AMH; genetic correlation; heritability; rima length; vulva measurements.

1 INTRODUCTION

The demand for animal protein is increasing and, consequently, producers need to become more efficient in livestock, being necessary to select genetically superior individuals. For the identification of superior animals, it is necessary to include the use of genetic evaluations of the cows, planning of directed matings, use of reproductive biotechnologies, being able to execute a greater selection pressure. Direct selection for fertility can become complex, due to the long period of evaluation of female performance and the low heritability of these traits (ZINK et al., 2011).

The identification of more easily measurable traits for fertility is necessary, so that the cows can be evaluated earlier (SILVA et al., 2005). Some studies indicate that the size of the external genitalia can be used as an indicator of fertility (MESQUITA et al., 2016; MACULAN et al., 2018; VASCONCELOS et al., 2020). Moreover, they have already proven that AGC is a good predictor of follicular reserve and ovarian functionality (GRIGOLETTO et al., 2020; IRELAND et al., 2008; MACULAN el al., 2018; MODINA et al., 2014).

Another parameter used as a fertility indicator is AMH, which is strongly associated with AFC (ALWARD e BOHLEN 2019; IRLANDA et al., 2011; GRIGOLETTO et al., 2020; MACULAN et al., 2018). Its advantage is the small variability over the various stages of the reproductive and estrous cycles of cattle, but with a high measurement cost (BALDRIGHI et al., 2014; BARRUSELLI et al 2015; MACULAN et al., 2018, ALWARD et. al., 2019).

Recent studies (MACULAN et al. 2018; MESQUITA et al. 2016; VASCONCELOS et al. 2020) have shown that AMH concentration and the size of reproductive structures (mainly the vulva) can be phenotypically correlated with AFC. However, Ribeiro et al. (2020) addressed that it has not been widely studied so far whether these traits have a genetic correlation and whether they are responsive to selection.

Understanding the correlations between fertility markers and external phenotypic traits can contribute to indirect selection. In this case, easy-to-measure and low-cost traits can be used as a selection criterion to maximize reproductive efficiency (MACULAN et al., 2018). This strategy would optimize the selection system, and consequently the producers would achieve a faster return on the investment, in addition to increasing the genetic gain, as it contributes to the reduction of generation interval.

Therefore, the aim of this study was to estimate the genetic parameters between AMH concentration, vulvar measurements and reproductive indices of the breeding program of

Zebu cattle (PMGZ) in Tabapuã cattle.

2 MATERIAL E METHODS

2.1 Location and Animals

Data was collected from 934 Tabapuã females aged between two and twenty-one years old. This database was generated in the collections carried out in 2013 and 2021, containing information on reproductive traits from nine farms located in the states of Rio de Janeiro (Fazenda Rodeio Gaúcho, Fazenda São Bento), Minas Gerais (Fazenda Chapadão and Fazenda Quatro Irmãs) and São Paulo (Fazenda Água Milagrosa, Fazenda Porto Seguro, Fazenda Córrego dos Dourados and Fazenda São José das Palmeiras).

The animals were raised in an extensive system and with mineral supplementation. The females had a known father and mother, which came from natural mating, artificial insemination or through embryos. The animals that obtained the measured measurements were not in heat at the time of collection, were not pregnant and were more than 30 days postpartum.

After database consistency, 800 animals were taken for analysis. Females were classified by parity, being divided into heifers n=315 (animals without any calving) and cows n=485 (animals with one or more calvings). All cows had known father and mother.

The zootechnical data file contained the following information on reproductive traits: rima length (RL), vulvar width (VW), Anti-Mullerian hormone (AMH) dosage and indices generated by the PMGZ, such as: reproductive efficiency (RE), age at first calving (AAFC), most probable producing ability (MPPA) and precocity, rusticity and survival (PRS).

2.2 Phenotypic measurements
2.2.1 External genital measurements

Vulvar measurements (rima length and vulvar width) were obtained using a digital caliper, according to the methodology described by Mesquita et al. (2016). The rima length (RIMA) was considered the length between the dorsal and ventral commissures of the vertical slit of the animal's vulva (Figure 1 A), whereas the vulvar width (VW) was obtained between the distances of the sides of the vulva from the midpoint of the rima, at an angle of 90 degrees (Figure 1 B).

Figure 1. Measurements of rima lenght and vulvar width
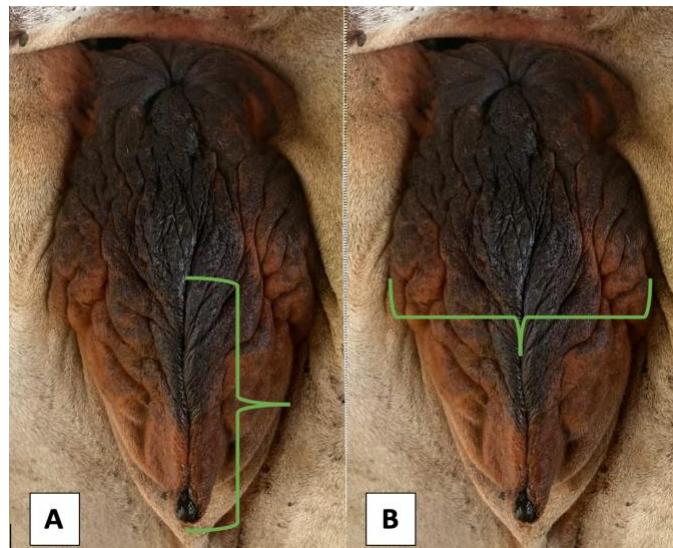
Legend: (A) rima length (A); vulvar width (B).

2.2.3 Evaluation of reproductive efficiency

Reproductive efficiency was estimated from the reproductive history of the females provided by the properties and indices provided by the breeding program of Zebu cattle (PMGZ), through the reproductive efficiency certificate. The variables studied and made available by the PMGZ were reproductive efficiency (RE), precocity, rusticity, and survival (PRS), most probable producing ability (MPPA) and age at first calving (AAFC), a technical indicator that reports the sow's age at its first birth.

The reproductive efficiency is obtained by the following formula:

$$ER = \frac{(n-1) \times 365}{D} \times 100$$

Where:

ER= reproductive efficiency;

n= numbers of calvings,

D= number of days between first and last calving

The RE expresses the cow's capacity to give birth regularly, considering that a standard cow is the one that had the first calving up to 36 months and then a calving every 365 days. In this way we would have an RE of 100%, that is, the closer to 100 this index is, the better the matrix will be.

The PRS is obtained by the following formula:

$$PRS = \frac{IDUP - 365}{NDE + 1}$$

Where:

PRS= biological index that combines traits of sexual precocity, fertility, maternal instinct, and degree of immunity to the offspring;

IDUP= age in days at the last calvin;

NDE= number of effective weanings;

The PRS combines traits such as sexual precocity, fertility, maternal instinct and the degree of immunity conferred on the offspring, for which all weaned offspring of the mother analyzed are considered (ABCZ file). The lower this index the better, as it demonstrates that throughout its life the cow has shown high sexual precocity and good maternal instinct.

The most probable producing ability is obtained by the following formula:

$$HMMP = 100,0 + \frac{n \times r}{1 + (n-1) \times r} \times (\overline{V} - 100,0)$$

Where:

HMMP= most probable producing ability

100.0= average of the calculated weight index in the herd, at the standard age of 205 days

r= 0.4, repeatability factor for the weight index calculated at weaning, according to specialized literature

V- average of weight indices calculated at the standard age of 205 days in the herd, relative to offspring of the same cow

The MPPA indicates the ability of the cow to wean the calf. In this case, the milking capacity and maternal instinct of the cow are very important. The higher this index the better.

2.2.4 AMH dosage

AMH quantification was performed in 845 animals, 323 nulliparous, and 522 primiparous and multiparous. Blood samples were collected by coccygeal venipuncture in 10 mL vacuum tubes (BD VACUTAINER®, São Paulo, BR). Serum was obtained after centrifugation at 3,000g for 15 minutes and kept frozen at -20°C for later analysis. AMH serum concentrations were determined by ELISA (Enzyme-Linked Immunosorbent Assay kit; Ansh Labs, Webster, Texas, USA), previously validated for cattle (IRELANDA et al.,

2008). One sample was conducted with a sensitivity of 0.011 ng/mL, being intra and inter assay with CV% 2.2%. The assays were carried out at the IgAc laboratory (GENESE Institute of Scientific Investigations, São Paulo, BR).

2.3 Statistical analyzes

The consistency of the database was carried out using the SAS statistical program (SAS, 2010), and after the exploratory analysis, discrepant information was removed (outliers), according to the methodology described by Freitas et al. (2008).

Contemporary groups (CG) were formed by the following variables: year of birth, farm and parity, and those CG that had less than three observations were eliminated. In order to identify which environmental effects significantly influenced ($P<0.05$) the evaluated traits, analyzes of variance were carried out using the least squares method, using the GLM procedure of the SAS software (SAS, 2010) considering a model with fixed effects of farm, parity, year of birth.

The animal model used for all traits studied included, as systematic effects, the contemporary group, the age class, and the live weight class of the animal at the time of measurements, and as the random effect, the animal itself.

The statistical model used, in matrix form, is described as follows:

$$y = \mathbf{X}\beta + \mathbf{Z}\alpha + e \ (1.0)$$

where:

y = vector of dependent variables;

X = fixed effects incidence matrix;

β = vector of fixed effects;

Z = random effects incidence matrices;

α = vector of direct additive genetic effects;

e = vector of residual random errors associated with observations.

The genetic parameters for vulvar traits (RIMA and VW), AMH and reproductive indices were estimated through two-trait Bayesian analyses, GIBBSF90 software (MISZTAL et al., 2002), as they are continuous traits.

For all traits, later estimates were obtained using the executable POSTGIBBSF90 (MISZTAL et al., 2002). The samples of the (co)variance components were obtained from

chains of one million iterations, in which the initial 100,000 iterations were discarded (burn-in) and the samples were removed every 5 cycles (thinning interval).

To perform the convergence tests, the Geweke, Heidel & Ralftery and Raftery & Lewis methods were used using the "BOA" package of the R program (the R v. 3.5.0 (R Core Team, 2018).

3 RESULTS

The mean value observed for AMH in the present study showed high variation in the population studied, with a coefficient of variation (CV) above 60. The mean value obtained for RIMA and VW did not show a large variation (Table 1).

Table 1 - Descriptive statistics of rima length (RIMA), vulvar width (VW), Anti-Mullerian Hormone (AMH), age and weight at the time of measurements of Tabapuã females.

Trait	N	Mean ± SD	Minimum	Maximum	CV
Age (days)	800	2164.7 ± 1489.2	310.0	6127.0	68.3
Weight (Kg)	800	447.7 ± 107.3	192.0	762.0	23.9
RIMA (cm)	800	8.8 ± 1.8	3.3	14.8	21.1
VW (cm)	800	7.6 ± 2.02	3.0	17.0	26.6
AMH (ng/mL)	442	0.8 ± 0.5	0.0	4.5	68.3

N = Number of observations; SD = standard deviation, CV = coefficient of variation.

The weight and age of the animals obtained presented a great variation 447.7±107.3, 2164.7±1489.2 respectively, presenting a CV of 23.9 and 68.3, showing the heterogeneity of the analyzed database (Table 1).

Table 2 – Descriptive statistics of reproductive efficiency indices of Tabapuã females provided by PMGZ.

Trait	N	Mean ± SD	Minimum	Maximum	CV
MPPA	492	100.0 ± 5.9	77.1	136.5	5.9
PRS	470	504.7 ± 181.1	61.5	1590.0	35.8
AAFC	586	1189.7 ± 353.4	688.0	3205.0	29.7
ER	432	74.9 ± 19.6	15.8	111.1	26.2

MPPA = most probable producing ability; PRS = precocity, rusticity and survival; AAFC = age at first calving; RE = reproductive efficiency.

There was considerable variation between the reproductive efficiency indices provided by the PMGZ. The mean value of PRS was 504.7±181.1 and that of RE was 74.9±19.6, with coefficients of variation above 35 (Table 2). These indices have a smaller number of data, as they are generated from the reproductive life of the animal and are updated every six months.

Table 3 - Additive genetic variance, credibility interval, heritability's and credibility intervals of reproductive traits and biometric measurements in Tabapuã females.

Parameters	Vga	CI	h^2	CI h^2
RIMA	0.690	0.310 - 1.080	0.32	0.158 - 0.508
VW	0.392	0.075 -0.719	0.20	0.043 - 0.376
AMH	0.106	0.023 - 0.192	0.16	0.087 - 0.586
MPPA	34364	763.30 - 84310	0.88	0.005 - 1.000
PRS	2569.1	262.60 - 5689.0	0.15	0.009 - 1.000
AAFC	7222.2	349.80 - 17270	0.28	0.000 - 0.029
ER	174.30	6.392 - 386.80	0.15	0.006 - 0.346

Additive genetic variance, credibility interval, heritabilities and credibility intervals of reproductive traits and biometric measurements in Tabapuã females, RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; AAFC = age at first calving; MPPA = most probable producing ability; PRS = precocity, rusticity and survival; RE = reproductive efficiency.

High heritability values were found for the trait: HMMP (0.88) and moderate values for: RIMA (0.33); VW (0.20); AAFC (0.28); and their respective credibility intervals are represented in the table 3. For these traits, we can say that there is a correlation between the

phenotype and the genetic value of the individual, with selection being more responsive. Extrapolation to other populations depends on how similar they are to the original genetic structures, measurement precision, environmental conditions, among other factors.

Table 4 – Genetic (above diagonal) and phenotypic correlations (below diagonal) between biometric traits, reproductive traits and reproductive indices evaluated in Tabapuã females. Heritability's are diagonally in bold.

	RIMA	VW	AMH	AAFC	MPPA	PRS	RE
RIMA	**0.32**	0.50	-0.23	0.12	0.35	0.14	-0.57
VW	0.49**	**0.20**	-0.26	0.48	0.03	0.19	-0.31
AMH	-0.09	-0.07	**0.16**	0.04	-0.11	-0.29	0.19
AAFC	-0.08	0.07	0.00	**0.28**	0.13	0.91	0.12
MPPA	-0.03	-0.09	0.00	-0.07	**0.88**	-0.77	-0.31
PRS	-0.04	-0.02	0.02	0.62**	0.01	**0.15**	0.20
RE	0.00	0.08	0.07	-0.03	-0.03	-0.35**	**0.15**

*$P < 0.005$ e **$P < 0.0001$. RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; AAFC = age at first calving; MPPA = most probable producing ability; PRS = precocity, rusticity and survival; RE = reproductive efficiency.

For traits related to reproductive aspects, a moderate phenotypic correlation was observed between VW and RIMA (0.49) (Table 3). For the other reproductive traits there was low or no correlation for the present study.

Table 5– Additive genetic covariance, its credibility intervals and genetic correlations between reproductive traits in Tabapuã females.

Correlations	COVg	CI	GC
RIMA x PRS	0.045	-0.806 – 0.999	0.14
RIMA x VW	0.548	0.092 – 1.000	0.50
RIMA x AMH	-0.155	-0.983 – 1.000	-0.23
RIMA x MPPA	0.376	-0.731 – 1.000	0.35
RIMA x AAFC	0.146	-0.919 – 1.000	0.12
RIMA x ER	-0.258	-0.999 – 0.843	-0.57
VW x AMH	-0.271	-0.100 —0.939	-0.26
VW x MPPA	0.041	-0.471 – 0.635	0.03
VW x PRS	0.204	-0.969 – 1.000	0.19
VW x AAFC	0.515	-0.594 – 1.000	0.48
VW x ER	0.392	-0.725 – 1.000	0.37
AMH x MPPA	-0.067	-0.992 – 1.000	-0.11
AMH x PRS	-0.280	-1.000 – 0.839	-0.29
AMH x AAFC	0.025	-1.000 – 0.997	0.04
AMH x ER	0.175	-0.910 – 1.000	0.19

COVg= mean of additive genetic covariance, CI= credibility interval, GC= genetic correlation, RIMA = rima length; VW = vulvar width; AMH = Anti-Mullerian Hormone; AAFC = age at first calving; MPPA = most probable producing ability; PRS = precocity, rusticity and survival; RE = reproductive efficiency

With regard to genetic correlations of reproductive traits, a moderate correlation was observed between RIMA and VW (0.50) so if selected for big RIMA would select for big VW.

However, when looking at the credibility intervals of the other analyses, we can say that there are no genetic correlations between the traits (Table 5). Thus, further studies should be conducted to confirm these relationships between these traits.

With the results obtained in this study, it is concluded that these traits are independent, that is, selecting one will not have a correlated response in another, these results could be because of the numbers of data studied. So, more studies are necessary.

4 DISCUSSION

Mean rima length (8.85±1.8) and vulva width (7.6±2.02) were similar to those found by Maculan et al. (2018) and Ribeiro et al. (2020), with the same breed. It is believed that the similarity is due to the breed studied and the similarity of the age group of the cows between the databases. Variation coefficients were also of low magnitude for these traits.

The variation observed in the concentration of AMH hormone (0.01 to 4.51 ng/mL) was similar to that observed by Maculan et al. (2018) (0.014 – 4.516 ng/mL), evaluating Tabapuã females. For Nellore heifers, Grigolleto et al. (2020) found a mean of 1.10±0.36, slightly higher than that verified in the present study. According to Batista et al. (2014), values of this hormone may vary between breeds, and Baruselli et al. (2015) stated that its concentration may have a small drop throughout life, which may explain the higher concentration obtained by Grigolleto et al. (2020).

The reproductive efficiency indices provided by the PMGZ also varied a lot, which can be explained by the great heterogeneity of the database used and by the age range of the animals, as these indices are generated from the reproductive life of the cow and are updated every six months with the fertility data of each female. The cows have a good ability to wean calves, with a mean of 100.0 ± 5.9 in the MPPA and good RE 74.9 ± 19.6, showing a good ability to calve regularly but do not have a high sexual precocity, fertility and maternal instinct, with a PRS mean of 504.7 ± 181.1.

For traits related to reproductive aspects, a moderate phenotypic correlation was observed between VW and RIMA (0.49) (Table 3). Analyzing 591 Tabapuã females, Maculan et al. (2018), estimated a divergent in relation to the correlation between RIMA and VW (0.09). These results may be due to the database being similar in relation to the origin and breed of the animals, but they present a smaller number of cows evaluated, leading to some divergences between the results (Table 3). The other phenotypic correlations are not relevant because they are not repeated in the genetic correlations, being traits greatly influenced by the environment.

The heritability obtained for AMH found was low (0.16), different from those found by Grigoletto et al. (2020), Gobikrushanth, et al. (2018) and Nawaz et al. (2018) including genomic information which observed a moderate to high heritability, 0.28, 0.36 and 0.46 respectively. This difference in magnitude may be due to variations in terms of genetic values and phenotypes between populations, due to the smaller database used by Grigoletto et al. (2020) (n=289) and by Gobikrushanth et al. (2018) (n=198). It is interesting to carry out further studies to better understand the genetic parameters involving AMH.

In this study, it was verified that the genetic correlations between RIMA and VW (0.50) are positive and moderate, selecting for the increase of one of them will cause a correlated response for the other trait in the same direction.

According to the results obtained in this study, we conclude that these traits are independent, that is, the selection of one will not cause any correlated response in the other.

One of the reasons for this large credibility interval found may be due to the number of data studied. Thus, further research involving these characteristics should be conducted.

5 CONCLUSIONS

In this study, moderate heritabilities was found for rima length, age at first calving, most probable producing ability and vulvar width, showing that they respond to direct selection. However, it was observed that there is no genetic correlation between the characteristics studied. So further research involving these characteristics should be conducted.

REFERENCES

ALWARD, K. J.; BOHLEN, J. F. Overview of Anti-Müllerian hormone (AMH) and association with fertility in female cattle. **Reproduction in Domestic Animals (Wiley)**, v. 55, n. 1, p. 3-10, 2019

BALDRIGHI, J. M. *et al*. Anti-Müllerian hormone concentration and antral ovarian follicle population in murrah heifers compared to holstein and gyr kept under the same management. **Reproduction in Domestic Animals**, v. 49, n. 6, p. 1015–1020, 2014.

BARUSELLI, P. S. *et al*. Relationship between follicle population, AMH concentration and fertility in cattle. **Animal Reproduction**, v. 12, n. 3, p. 487-497, 2015.

BATISTA, E. O. S. *et al*. Plasma Antimullerian Hormone as a Predictor of Ovarian Antral Follicular Population in *Bos indicus* (Nelore) and *Bos taurus* (Holstein) Heifers. **Reproduction in Domestic Animals**, v. 49, n. 3, p. 448-452, 2014.

FREITAS, A. R. et al. Técnicas de análises exploratórias em dados de cultivares de alfafa. **Revista Brasileira de Zootecnia**, Viçosa, MG, v. 37, n. 9, p. 1532- 1532, 2008.

GOBIKRUSHANTH M., *et al*. The relationship between serum anti-Müllerian hormone concentrations and fertility, and genome-wide associations for anti-Müllerian hormone in Holstein cows. **Journal of Dairy Science** v. 101, n. 8, p. 7563-7574, 2018.

GRIGOLETTO, L. *et al*. Genetic Parameters and Genome-Wide Association Studies for Anti-Müllerian Hormone Levels and Antral Follicle Populations Measured After Estrus Synchronization in Nellore Cattle. **Animals**, v. 10, n. 1185, p. 1-15, 2020.

IRELAND, J. L. H. *et al*. Antral Follicle Count Reliably Predicts Number of Morphologically Healthy Oocytes and Follicles in Ovaries of Young Adult Cattle. **Biology of Reproduction**, v. 79, n. 6, p. 1219-1225, 2008.

IRELAND, J.J. *et al*. Does size matter in females? An overview of the impact of the high variation in the ovarian reserve on ovarian function and fertility, utility of anti-Mullerian

hormone as a diagnostic marker for fertility and causes of variation in the ovarian reserve in cattle. **Reproduction Fertility and Development**, v. 23, p. 1–14, 2011.

MACULAN, R. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Animal Reproduction Science**, n. 189, p. 84-92, 2018.

MESQUITA, N. F. *et al*. Anti-Müllerian Hormone (AMH), antral follicle count (AFC), external morphometrics and fertility in Tabapuã cows. **Journal of Reproduction and Development**, v. 62, n. 6, p. 1-4, 2016.

MODINA, S. *et al*. Reductions in the number of mid-sied antral follicle are associated with markers of premature ovarian senescence in dairy cows. **Reproduction, Fertility and Development**, v. 26, n. 2, p. 235-244, 2014.

NAWAZ M. Y. *et al*. Genomic heritability and genome-wide association analysis of anti-Müllerian hormone in Holstein dairy heifers. **Journal of Dairy Science,** v. 101, n. 9, p. 8063-8075, 2018.

RIBEIRO, Y. P. **Parâmetros genéticos para características indicadoras de fertilidade em fêmeas da raça Tabapuã**. 2020. 28 p. Trabalho de conclusão de curso (Bacharelado em Zootecnia) - Universidade Federal de Lavras, Lavras, 2020.

SILVA, J. A. *et al*. Estudo genético da precocidade sexual de novilhas em um rebanho Nelore. **Revista Brasileira de Zootecnia,** v. 34, n. 5, p. 1568-1572, 2005.

VASCONCELOS, G. L. *et al*. Effects of vulvar width and antral follicle count on oocyte quality, in vitroembryo production and pregnancy rate in Bos taurus taurus and Bos taurus indicus cow. **Animal Reproduction Science,** v. 217, n. 106357, 2020.

ZINK, V. *et al*. Genetic parameters for female fertility, locomotion, body condition score, and linear type traits in Czech Holstein cattle. **Journal Dairy Science**, v. 94, n. 10, p. 5176- 5182, 2011.v. 94, p. 5176- 5182, 2011.